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THE PLANT DISEASE REPORTER

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PLANT DISEASE EPIDEMICS
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IDENTIFICATION SECTION

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The Plant Disease Reporter is issued as a service to plant pathologists throughout the United States. It contains reports, summaries, observations, and comments submitted voluntarily by qualified observers. These reports often are in the form of suggestions, queries, and opinions, frequently purely tentative, offered for consideration or discussion rather than as matters of established fact. In accepting and publishing this material the Plant Disease Epidemics and Identification Section serves merely as an informational clearing house. It does not assume responsibility for the subject matter.

THE PLANT DISEASE REPORTER

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PLANT DISEASE EPIDEMICS AND IDENTIFICATION SECTION

Horticultural Crops Research Branch — Plant Industry Station, Beltsville, Maryland
Volume 40 — July 15, 1956 — Number 7

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INFLUENCE OF CAULIFLOWER MOSAIC AND TURNIP MOSAIC VIRUSES ON YIELDS OF CABBAGE IN NEW YORK STATE[1]

J. J. Natti

SUMMARY

In experimental plots, yields from cabbage plants inoculated in the seedbed, or early in the season, with a strain of cauliflower mosaic and of turnip mosaic viruses ranged from 50 to 75 percent less than those from virus-free plants or plants inoculated late in the season. Infection of plants in the seedling stage resulted in almost complete failure of the crop. Late season infections had no noticeable influence on yields.

In commercial plantings, virus diseases do not appear to reduce yields even though a high percentage of the plants may be infected at harvest-time. The results of these tests indicated that plants in commercial fields do not become infected until late in the season. The absence or very low incidence of virus-infected plants early in the season in commercial plantings appears to be related to the availability of inoculum. Under natural conditions, the viruses must be harbored over winter in biennial or perennial hosts which occur either in low population, or are not attractive to the insects which transmit these viruses.

INTRODUCTION

Virus diseases of cabbage are not considered of economic importance in western New York State. Evidence has been obtained, however, that indicates that virus diseases of cabbage may at times be of considerable importance. In 1951 at Geneva, New York, 111 of 141 cabbage plants randomly selected from a commercial field at harvest-time were found to be infected with one or two viruses. Because of this high incidence of virus-infected plants under field conditions, experiments were conducted to determine whether virus diseases of cabbage were of sufficient importance, on the basis of yield reduction, to warrant the development of control measures. The results of these tests indicated that yields from plants infected in the seedbed or in the field soon after transplanting were significantly less than those from plants infected later in the season or from plants which remained virus-free.

MATERIALS AND METHODS

1952 Season: Cabbage plants of the varieties Improved Wisconsin Ballhead, Klitgord's Selected Danish, Robson's Seneca Danish, Klitgord's Yellows Resistant Early Kraut, Resistant Glory, Wisconsin All Seasons, Geneva Danish lines 107, 140 and 153, and Geneva kraut lines 88 and 89 were grown in greenhouse flats. On July 8, at which time the seedlings were at the 4-leaf stage of growth, one-third of the plants of each variety and breeding line were inoculated by lightly rubbing the upper surface of one leaf of each plant with a pad of cheesecloth which had been dipped in juice expressed from leaves of cabbage plants known to be infected with a strain of turnip mosaic virus (Marmor brassicae Holmes) and of cauliflower mosaic virus (M. cruciferarum Holmes). Powdered carborundum was used as an abrasive. All plants were removed to cold frames on July 12. On July 28 they were transplanted into field plots. The planting consisted of 4 replications of paired rows of virus-free and virus-infected plants with 25 plants to a row. The virus-free plants were planted before the virus-infected in order to avoid the mechanical transmission of the viruses to the virus-free plants during the planting operations. Another planting of virus-free plants was made in the same field but was separated from the plot containing the virus-infected plants by a planting of broccoli about 60 feet wide. Cultural practices were the same in each plot.

Insecticide sprays were applied to all plots at 10-day intervals beginning 18 days after transplanting.

The heads were harvested during the first week of November. Yield data were obtained by determining the total number and weight of all heads, marketable or otherwise, in each row.

1953 Season: Cabbage plants of the varieties Donk's Danish, Improved Wisconsin Ballhead, Harris' Danish, Robson's Seneca Danish, Klitgord's Yellows Resistant Early Kraut, Marion Market, Reed's Improved Glory, Wisconsin All Seasons, the breeding lines Cornell Danish

[1]Approved by the Director for publication as Journal Paper No. 1040, New York State Agricultural Experiment Station, Geneva, New York.

66E (now Empire Danish), Geneva Danish 55G-49, Geneva kraut 6950M, and Wisconsin mosaic-resistant Danish (obtained from Dr. Pound) were grown in greenhouse flats. On July 1, when the plants were at the 4-leaf stage of development, one-half of the plants of each variety and breeding line were inoculated with a mixed inoculum of turnip mosaic and cauliflower mosaic viruses, as previously described. All plants were transferred to cold frames 3 days later. On July 22 the plants were hand-planted into field plots. The planting consisted of 6 replications of paired rows of virus-infected and virus-free plants of each variety and breeding line. Each row contained 20 plants.

All plants were sprayed at weekly intervals for a total of 5 sprays beginning 18 days after transplanting.

The plants were harvested during the first week of November. Yield data were obtained by determining the total number and weight of all heads in each row.

1954 Season: Plants of the cabbage varieties Marion Market and Reed's Improved Glory were started in the greenhouse on May 19. Two-thirds of the plants of each variety were maintained virus-free in a greenhouse section. The remaining plants of each variety were inoculated with a strain of turnip mosaic and cauliflower mosaic viruses by means of cabbage aphids (Brevicoryne brassicae Linn.) when the majority of the plants were at the 4-leaf stage of development. After a 3-day feeding period the aphids were removed by fumigating the greenhouse with nicotine sulfate. All plants were then transferred to cold frames. On July 6 the plants were hand-planted into field plots. The planting consisted of one row of infected plants and two rows of virus-free plants of each variety randomized within each of 6 replications. Each row contained 16 plants. The plants in one of the virus-free rows of each variety in each replicate were sprayed with a mixture of nicotine sulfate and parathion emulsion every 3 days for a total of 7 applications to keep these plants free from insects. The other plants in the plots were not sprayed with insecticides until 21 days after they were set into the field. Insecticide sprays were then applied to the entire planting at about 10-day intervals until the end of the season.

The plants were harvested on October 21. Yield data were obtained by determining the total number and weight of all heads in each row.

RESULTS

The plants which were experimentally inoculated with the viruses in the greenhouse were systemically invaded by transplanting time. The earliest symptoms consisted of vein clearing and mottling. As infection progressed, the lamina of the leaves became coarsely mottled or savoyed, and development of leaf bloom was suppressed in scattered areas or over the entire surface of the leaves. Several weeks after transplanting, the older leaves became necrotic and abscised. This type of defoliation continued for the entire season.

In the tests conducted in 1952 and 1953, plants which were virus-free at the time they were set into the field began to develop symptoms about 3 weeks after transplanting. The symptoms were similar in type but were less severe than those of plants experimentally inoculated in the greenhouse. In the field, the plants became infected by viruses transmitted by aphids migrating from the experimentally inoculated plants.

In the 1954 test, disease symptoms of plants inoculated by aphids in the greenhouse were similar to those of plants in the 1952 and 1953 tests in which inoculations were made by rubbing the leaves with juice containing the viruses. In 1954, most of the plants which were virus-free at transplanting time and were not sprayed with insecticides until 21 days later had developed symptoms within 3 weeks. Most of the plants which were virus-free at transplanting time and were sprayed every 3 days for a total of 21 days began to develop symptoms 40 to 50 days after transplanting. Symptoms were present in some plants before 40 days and in others after 50 days. At harvest time all plants were infected.

In all tests, yields from plants experimentally inoculated when at a seedling stage before transplanting were significantly less than those from plants which became infected in the field (Tables 1, 2, 3) with the single exception of plants of the Wisconsin mosaic resistant Danish line (Table 2). Leaf necrosis and defoliation appeared to be less severe in this line than in the commercial varieties and other lines. In the 1952 test, none of the plants inoculated in the greenhouse produced a marketable head. Defoliation was so severe that only a few mature heads were formed. In the 1953 and 1954 tests, some of the plants inoculated before transplanting produced heads of marketable size, but the market value of many heads was destroyed by necrotic spots in the leaves. In practical terms, the results showed that seedbed infection of cabbage resulted in the complete failure of the crop during three successive years.

Table 1. Yields of cabbage from plants inoculated at different stages of maturity with a strain of turnip mosaic and cauliflower mosaic viruses[a].

Variety	Plants inoculated		Virus-free plants[d]
	In seedbed[b]	In field[c]	
	Tons per acre		
Improved Wisconsin Ballhead	2.9	5.6	13.3
Klitgord's Selected Danish	4.0	7.0	13.3
Robson's Seneca Danish	6.3	8.5	14.1
Yellows Resistant Early Kraut	4.4	10.1	17.7
Resistant Glory	4.0	8.6	19.2
Wisconsin All Seasons	3.2	8.1	17.5
Geneva Danish 107	2.5	8.0	14.3
Geneva Danish 140	5.1	8.4	13.0
Geneva Danish 153	1.5	3.5	13.7
Geneva kraut 88	5.5	8.6	19.5
Geneva kraut 89	4.1	7.2	20.0
Least difference required for significance (19:1)		2.1	—
(99:1)		2.7	—

[a]Yield data obtained from 4 replications of 25 plants per replicate.
[b]Plants experimentally inoculated in the seedbed 27 days before transplanting to field.
[c]Plants naturally inoculated in the field.
[d]Plants grown in isolated plot 60 feet from virus plot. Data not included in statistical analysis. Most of these plants became infected late in the season.

Table 2. Yields of cabbage from plants inoculated at different stages of maturity with a strain of turnip mosaic and cauliflower mosaic viruses, 1953[a].

Variety	Plants inoculated		Virus-free plants[d]
	In seedbed[b]	In field[c]	
	Tons per acre		
Donk's Danish	6.1	9.5	18.9
Harris' Danish	4.8	8.1	16.8
Improved Wisconsin Ballhead	4.9	7.7	16.5
Robson's Seneca Danish	5.2	8.9	19.2
Marion Market	6.9	9.2	20.6
Reed's Improved Glory	5.6	9.4	23.7
Wisconsin All Seasons	7.6	10.4	19.4
Yellows Resistant Early Kraut	6.0	10.0	20.7
Cornell Danish 66E	6.3	9.3	19.1
Geneva Danish 55G-49	4.4	6.0	14.2
Geneva kraut 6950M	5.3	8.2	21.9
Wisconsin mosaic-resistant Danish	5.3	6.1	12.3
Least difference required for significance (19:1)		1.1	—
(99:1)		1.4	—

[a]Yield data obtained from 6 replications of 20 plants per replicate.
[b]Plants experimentally inoculated in greenhouse 21 days before transplanting to field.
[c]Plants naturally inoculated within 3 weeks after transplanting.
[d]Yield from separate plot in which plants remained virus-free or became infected late in the season. Data not included in statistical analysis.

Table 3. Yields from cabbage plants inoculated at different stages of maturity with a strain of turnip mosaic and cauliflower mosaic viruses[a].

Variety	Seed bed inoc.[b]	Early field inoc.[c]	Late field inoc.[d]
	Tons per acre		
Marion Market	5.5	8.9	13.2
Reed's Improved Glory	8.2	10.9	16.4
Least difference required for significance (19:1)		0.6	
(99:1)		0.8	

a Yield data obtained from 6 replications of 16 plants per replicate.
b Plants inoculated in greenhouse by aphids.
c Plants naturally inoculated within 21 days of transplanting.
d Plants naturally inoculated later than 21 days after transplanting. Plants were sprayed with insecticide every 3 days for the first 21 days after transplanting.

Yields from plants which became infected soon after transplanting, although greater than those from plants infected before transplanting, were considerably less than those from plants infected later in the season. In the 1952 and 1953 tests, yields from plants infected within 3 weeks of transplanting averaged about 50 percent less than those of plants infected later in the season (Tables 1 and 2). This comparison is not valid from a statistical viewpoint since the plants which became infected late in the season were grown at a different location than those infected soon after transplanting. However, since all the plants were grown under identical cultural practices, and the yields obtained from the plants which became infected late in the season or which remained virus-free were representative of the average yields obtained by growers in this area, the comparison does indicate that field infection of cabbage early in the season can markedly reduce yields. In the 1954 test, yields from plants in plots in which infection was delayed by intensive application of insecticides averaged about 30 percent greater than those from plots in which the plants were infected soon after transplanting (Table 3). Some of this greater yield may be attributed to insect control, but since no build-up of insect population was observed in any of the plots, the differences in yield were due primarily to the stage of growth of the plants at time of invasion by the viruses.

DISCUSSION

The determination of the influence of virus infection on cabbage yields is complicated by the rapid spread of the virus in the field under conditions in which a supply of inoculum is readily available to the insect vectors. The spread of the viruses in the field can be delayed to some extent by the intensive application of insecticides, but the spread cannot be prevented. For this reason, it is difficult to conduct yield tests consisting of randomized plantings of virus-free and virus-infected cabbage plants. The methods employed in these tests to overcome this difficulty are open to criticism. In the 1952 and 1953 tests, plants were maintained virus-free until late in the season by growing them in plots isolated from the virus-infected plants. This method has the obvious disadvantage of resorting to comparisons in yield from plants grown at different locations. In the 1954 tests, plants in some plots were maintained virus-free for at least 3 weeks by the intensive application of insecticides while plants in unsprayed plots became infected soon after being transplanted. This method was not entirely satisfactory because some of the differences in yield may have been due to insect control as well as to delay in infection of the plants.

Despite the difficulties involved in conducting clear-cut tests on the influence of virus infection on cabbage yields, the results showed that virus disease of cabbage can be of consider-

able economic importance. Infection of the plants in the seedbed, or early in the season in the field, can result in the complete failure of the crop. Although virus diseases have not been observed to be sufficiently severe in commercial cabbage plantings to cause a noticeable reduction in yield, growers should be made aware of the potential seriousness of these diseases. It is possible that in certain years when conditions are favorable for the early season spread of crucifer viruses that entire cabbage plantings can be destroyed. To safeguard the production of cabbage, methods for control of virus diseases should be developed. The breeding of virus-resistant varieties appears to be the most promising method of control.

Virus diseases of cabbage in commercial plantings in western New York do not appear to have much effect on yields, even though a high percentage of the plants may be infected with one or two viruses at harvest-time. Under ordinary field conditions, therefore, the cabbage plants must not be invaded by the viruses until the plants are approaching or have attained their maxiumm growth. The results of the tests conducted on the influence of virus infection on yields of cabbage provide indirect evidence that the absence or low incidence of infected plants in commercial plantings early in the season is related to the availability of inoculum. This interpretation is based on the observations that in plots which contained experimentally infected plants, the viruses were spread to healthy plants within the same plot so rapidly that all plants were infected within 3 weeks of the time of transplanting; whereas, in plots containing only virus-free plants, the spread of the viruses did not take place until late in the season.

Under natural conditions the viruses must overwinter in biennial or perennial hosts which occur either in very low population or are not attractive to the insects which transmit the cabbage viruses. The low incidence of virus-infected cabbage plants in commercial plantings early in the season can be attributed to the inoculation of individual cabbage plants by stray viruliferous aphids migrating into the planting. The general occurrence of virus-infected plants late in the season can be attributed to the spread of the viruses within the planting from inoculum provided by the few originally infected cabbage plants.

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STRAWBERRY VIRUS TRANSMISSION BY GRAFTING EXCISED LEAVES

R. S. Bringhurst and Victor Voth

ABSTRACT

Strawberry viruses are readily transmitted by cleft grafting excised terminal leaflets from test plants in place of the terminal leaflets of indicator plants. Successful graft unions can be quickly detected and virus symptoms develop in from 2 to 5 weeks on alpine vesca. Leaves can be collected from the field and grafted immediately or stored in polyethylene for a month or more at 36° F before grafting.

A simple method of virus transfer is desirable in order to facilitate indexing and artificial infection of strawberry plants. Modifications of the stolon grafting method of Harris (1) are useful but the receptor and donor plants must both be growing vigorously and one or both must be producing stolons. This is not always possible if a plant is severely affected by the virus it carries.

An excised leaf grafting procedure we have experimented with may be helpful. The technique and preliminary results are described in this report.

MATERIALS AND METHODS

Commercial plants used included ten virus-infected California selections and seven that are presumed to be virus-free. Non-runnering alpine vesca seedlings and a runnering clone of Fragaria vesca obtained from N. W. Frazier were used as indicator plants.

The grafting material is a self-adhesive latex bandaging tape called "Sealtex". This is probably similar to the "crepe rubber" mentioned by Harris and Hildebrand (2).

The procedure is illustrated in Figure 1. Mature, fully formed leaves are better than immature leaves for both "stock" and "scion". The lateral leaflets of the scion leaf are removed and two-thirds or more of the terminal leaflet is severed in order to reduce transpiration. The scion petiole is trimmed with a razor blade to form a tapered wedge of 8 to 10 mm. The wedge should be thin at its tip. The scion leaf must be kept moist and should be placed in water while the stock petiole is prepared.

The terminal leaflet of the stock petiole is removed and the petiole is split equally between the lateral leaflets. The depth should be sufficient to accomodate the wedge of the scion petiole. The scion petiole is inserted in the cleft of the stock petiole and the union is tightly wrapped from the base upward. The tape can be conveniently handled in approximately 8 mm x 50 mm strips and can be protected from rapid deterioration by wrapping it with cellophane tape or coating it with a water-asphalt emulsion. It is imperative that the union be tightly wrapped. The self-adhesive latex can be stretched as the wrap is made thus binding the union securely. Thin dental rubber was successfully used in the same manner but it is not self-adhesive and the wrap must be carefully secured with cellophane tape. Electricians' tape might be used but it is much more difficult to wrap than the latex material.

After grafting, the plants must be protected from excessive heat and should receive water each day for several days. The success of the graft can be determined in less than one week. The union heals completely through the entire incision area in many instances (Figure 2).

RESULTS

OCTOPLOID/DIPLOID GRAFTS: Most of the preliminary attempts involved the grafting of leaves from virus-infected commercial selections to alpine vesca. Alpine stock plants include mature plants over 1 year old and new plants less than 3 months old.

The transmission results for the first 21 plants grafted are given in Table 1. Only four successfully grafted plants failed to develop symptoms after 2 months. Of these, two involved leaves from plants presumed to be virus-free. Many plants have been successfully grafted since and the results are similar.

The average time between grafting and symptoms compares favorably with that for transmission by stolon grafting. Young alpine seedlings developed symptoms more quickly than older alpine plants. The older plants had multiple crowns and symptoms were detected in the growth from the crowns that had the leaf grafts earlier than in crowns that had no grafts.

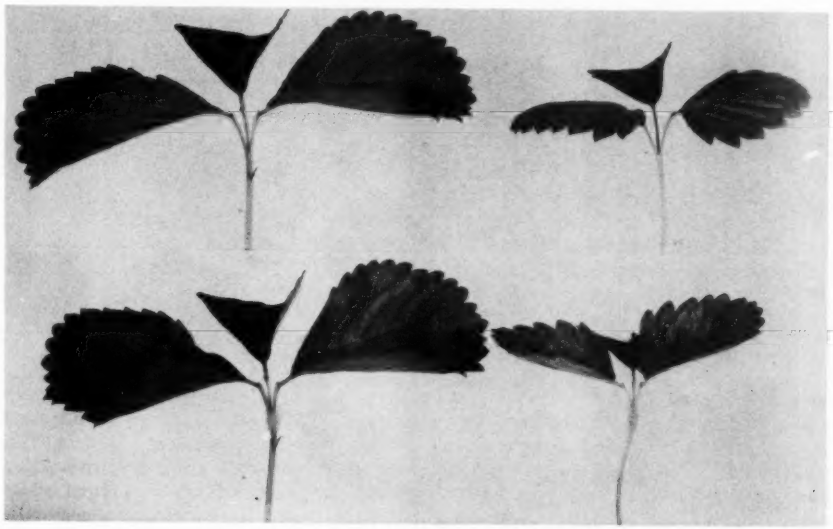

FIGURE 1. Illustration of grafting procedure; octoploid (Cupertino)/ octoploid (Lassen) left and octoploid (Cupertino)/diploid (Alpine vesca) right; before wrapping upper and wrap started lower.

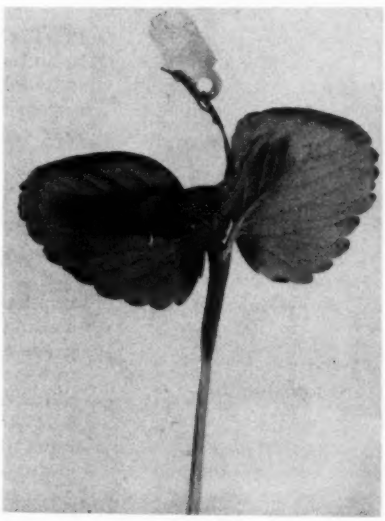

FIGURE 2.
Exposed healed union after 35 days, octoploid/octoploid.

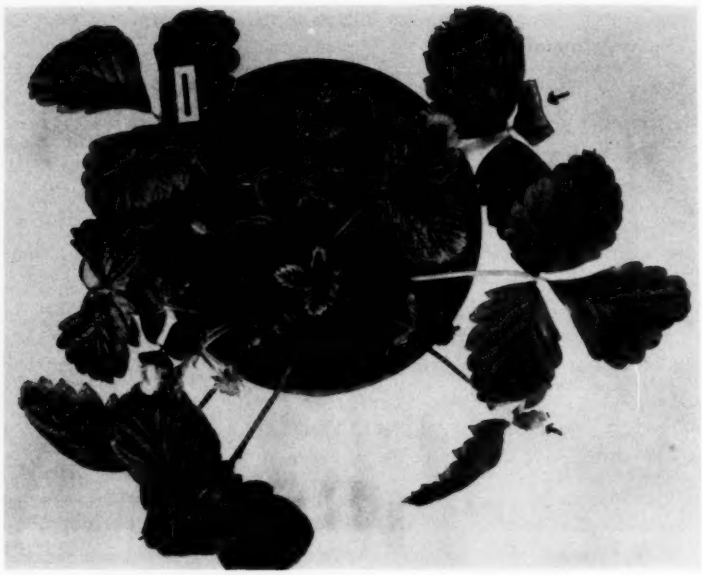

FIGURE 3. Symptoms in Alpine vesca 30 days after leaf graft of Cal 39.117-4. Note intact grafted terminal leaflets.

FIGURE 4. Severe symptoms in Alpine vesca 45 days after a single successful leaf graft of Cal 39.115-13.

Table 1. Indicator plant and days to first symptoms after excised leaf grafts.

Indicator	Number	Days to first symptoms	
		Average	Range
Mature alpine	14	25	17-34
Young alpine	5	15	14-16
Frazier vesca	3	35	21-50

Later results indicate that symptoms develop as rapidly in the Frazier vesca as in alpine.

A single successful leaf graft is apparently as effective in transmitting virus as multiple grafts (Fig. 3 and 4). As many as six were placed on a single plant but two to three are probably sufficient depending upon the operator. Grafts were made by two operators, one considerably more experienced than the other in handling the materials. The experienced operator successfully grafted 55 leaves in 62 attempts for 89 percent. The inexperienced operator successfully grafted 10 out of 24 for 42 percent, the first time he tried it. The operation is facilitated if the leaves from the octoploid plants do not have a petiole diameter more than one half greater than the vesca stock petioles. They should be matched as nearly as possible.

Successfully grafted leaves usually persist until after symptoms develop (Fig. 3). Lateral leaflets on the graft petiole frequently wither and die before the grafted terminal leaflet.

Leaves can be collected from the field for use in grafting. Leaves removed from a planting in San Jose, placed in polyethylene bags and carried over 100 miles to Davis were grafted the next day, and symptoms developed within 21 days.

Leaves can also be stored in polyethylene at 36°F for at least a month before grafting. Leaves collected in the field from four different selections have been successfully handled in this way. They were in as good condition for grafting after 1 month storage as they were fresh.

OCTOPLOID/OCTOPLOID GRAFTS: Leaves from commercial selections are easily grafted on plants of other commercial selections. In a preliminary experiment, the previously mentioned experienced operator successfully grafted 45 leaflets in 45 attempts. The inexperienced operator did almost as well with 17 successful in 18 attempts. One virus-free selection that has received leaves from a virus-infected selection may be developing symptoms.

DISCUSSION

The leaf grafting technique can be used to supplement grafting procedures now employed.

Systematic or random indexing of plants grown in isolation for certification might be accelerated. Leaves can be collected from the fields early in the season and grafted immediately into indicator plants. Results could be available in time to remove infected runner plants before stolons begin to form.

Weak plants may be indexed. Plants are often encountered in fruit plantings that are so weakened by apparent virus infection that they cannot be propagated. Leaves taken from such plants in the field were successfully used in experiments reported here. Such leaves were stored for 3 weeks before grafting in one experiment.

In breeding for virus tolerance, virus can be transmitted to selections by leaf grafting. A virus-tolerant clone, such as Shasta, can be infected with a combination of the viruses prevalent in the principal growing areas. The infected, tolerant plants may then be used as a source of leaves for inoculating selections to be tested. The test selections may then be grown in suitable comparison with the tolerant clone. This might yield information that will permit accurate prediction regarding the performance of the new selection relative to virus.

It may take longer to make a leaf graft than a stolon graft but the difference in the time involved is more than offset by the time saved in plant preparation. For leaf grafts, plants to be

indexed need not be propagated in the glasshouse and runners are not needed. A reasonably skilled operator can make 20 or more leaf grafts per hour.

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POMOLOGY
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MECHANICAL AIDS IN THE STUDY OF ERIOPHYID MITES IN RELATION TO YELLOW STREAK MOSAIC OF WHEAT[1]

Hurley Fellows[2]

Considerable study of Aceria tulipae has been made since the discovery that this eriophyid mite is a vector of the virus of yellow streak mosaic of wheat. The leaves upon which the mites feed and propagate soon become rolled, making examination for them and their eggs difficult. About 30-power magnification is required to see these mites plainly. The best instrument for their study is the binocular stereoscopic wide field microscope. Lacking this, any microscope may be utilized by using a 40 mm objective or an ordinary low power objective with the outer lens removed.

This paper describes various pieces of equipment that have been devised to make the study of mites easier. It is hoped that it may stimulate the development of other helpful devices.

Device No. 1 (Fig. 1A and B) was designed to keep the surface of the leaf under study in constant focus and also to aid in straightening a rolled or crinkled leaf. The one used by the author is shown separately in Figure 1A and in place on a microscope stage in Figure 1B. The base is of 1/8 x 3/4 inch brass with a length to suit the microscope stage on which it is being used. The uprights are formed from a piece of sheet copper bent up at both ends and soldered onto the base. The copper tube of 1/4-inch outside diameter is soldered to the holes in the two uprights. A convenient height above the base for the upper surface of the tube is about 7/8 to 1 inch. The device is attached to the stage with a pinch clamp with the tube directly under the objective so its upper surface is in focus. A leaf is grasped at its extremities and slowly pulled lengthwise over the tube.

Other simple devices may accomplish the same purpose. For example, Device No. 2 is made from No. 10 galvanized wire flattened at one end, clamped to the stage, and bent so that it is horizontal under the objective as shown in Figure 2.

An infested leaf specimen may be held in focus under the lens so that both hands are free by attaching the leaf to the raised side rails shown in Figure 2 with hair curlers, which are small aluminum spring clamps. The leaf is stretched across either Device No. 1 or Device No. 2. The hair curlers, shown in Figure 2, are light and they grasp the leaf firmly without injuring the tissue. The side rails are of sheet metal, 24 or 26 gauge, 1/4 inch wide, and raised about 1/8 inch from the stage. They are bent in such a way that they are held on the stage by friction and are easily removed.

One may desire to examine a leaf while still on the plant, with the leaf held firmly, preferably in a horizontal position. Figure 3 illustrates Device No. 3 used for that purpose, made from an inch long piece of No. 12 or 14 gauge galvanized wire. One end of the wire is bent to form a rectangle about 1/2 x 3/4 inch and the plane of the rectangle is bent at a right angle with the remainder of the wire, which is inserted in the soil. The leaf under examination rests on the rectangle and is fastened in place with hair curlers. The leaf should be as nearly level as possible so that all portions of it will be in focus.

Device No. 4 (Fig. 4 and 5) is a microscope tube with the lens so arranged that it may be swung over potted plants and focused for the study of leaf surfaces in place. It may be used in connection with Device No. 3. An extension binocular might be preferable to Device No. 4, but it is less versatile, heavier, and more expensive. The base is the bottom of a round ring stand with a 1/2-inch cold rolled steel upright 20 inches high (see Fig. 4). The rods and pinion are removed from an old microscope tube to reduce weight. The end of another 1/2-inch rod is held in the jaws of a laboratory clamp as shown in Fig. 4. These jaws should be widened for a firmer clasping action by welding to their inner surfaces two pieces of heavy sheet metal about 1 inch wide, first bent to fit snugly on the surface of the tube. The jaws of the clamp hold the microscope tube at right angles to the rod (Fig. 4).

Vertical and horizontal movements are provided for in the following manner (Fig. 4 and 5): The sides of two 1/2-inch couplers are welded together at their central point with their axes at 90°. The flat side of a 3/4-inch common washer with an outside diameter of 2 inches is centered and welded to form a flange on a third coupler and the surface of the washer is turned in a lathe

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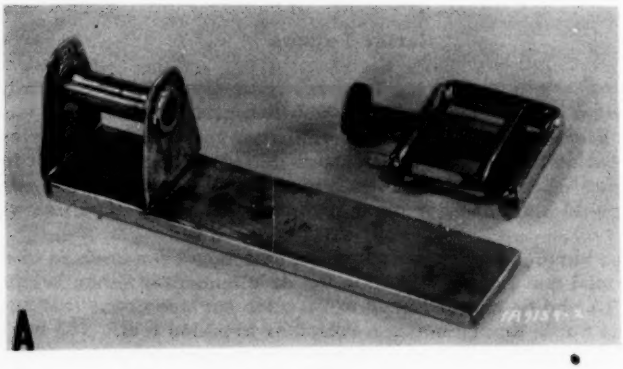

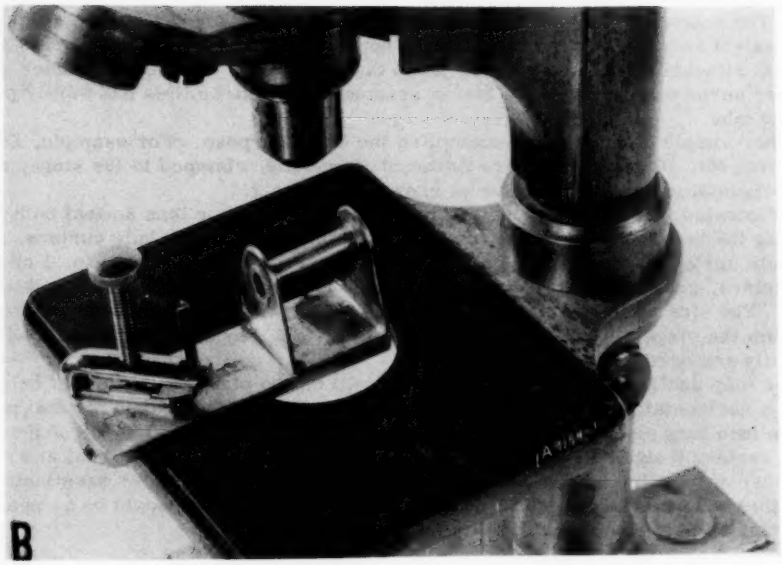

FIGURE 1, A. and B. A device fastened with a hose clamp to the stage of a microscope to keep a wheat leaf surface in constant focus while being moved laterally. This device also aids in unfolding a curled leaf.

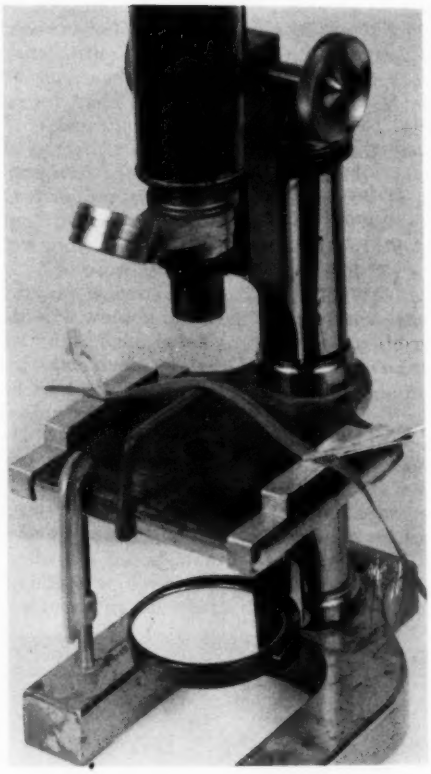

FIGURE 2. Raised side rails of sheet metal permit the holding of a severed wheat leaf in focus. The clamps, made of aluminum, are hair curlers.

FIGURE 3. A leaf on a growing wheat plant may be held firmly in place for microscopic examination by means of a bent wire and small aluminum clamps.

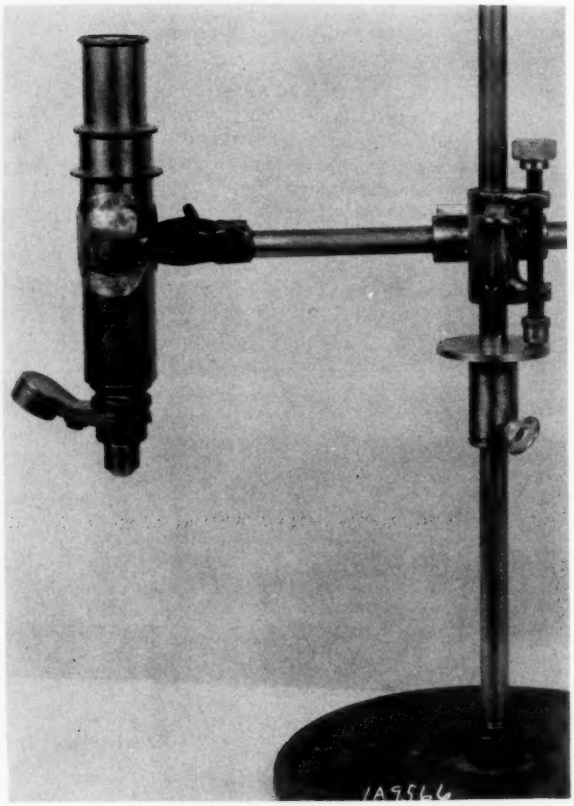

FIGURE 4. A simple inexpensive extension microscope may be made from materials available in most laboratories. It may be swung horizontally and has fine focusing adjustment.

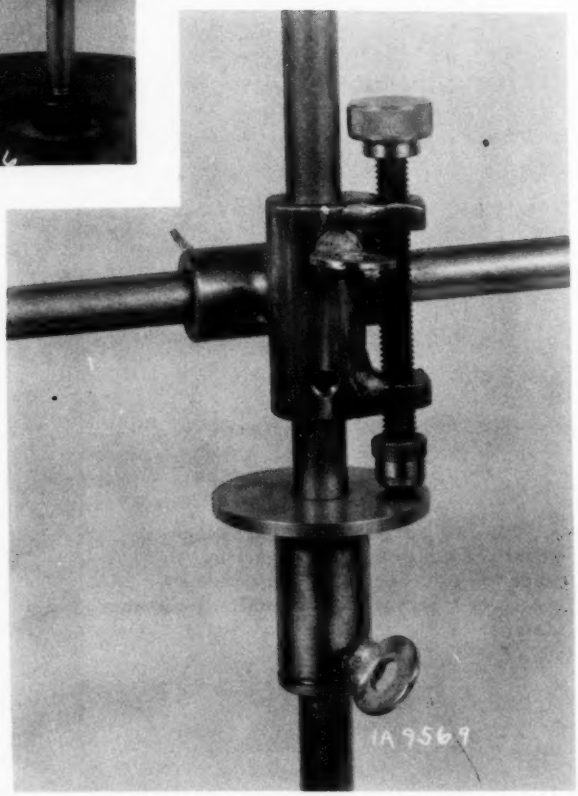

FIGURE 5. Details of the focusing portion of the improvised extension microscope.

after welding to make it smooth and its surface perpendicular to the coupler (Fig. 5).

Couplers may be purchased in most stores handling small homecraft machinery. The threaded holes in their side walls fit a round-headed stove bolt 3/16-inch in diameter. The heads of these bolts are provided with a wing for finger turning by welding the edge of a common 1/4-inch washer into the screwdriver slot. These winged bolts are then placed in the couplers, one for each coupler.

When these parts are assembled, the coupler with the large washer flange is first slid over the upright rod with hte washer flange up. It may be placed at any convenient point and held in place with the winged bolt previously described. Then one of the welded pair of couplers is slid over the upright until it rests on the flanged coupler. The rod holding the microscope tube is then slid into the other coupler of the welded pair. This provides for everything, except for the slow movement necessary for accurate focusing.

The slow movement is fixed to the welded couplers rather than to the microscope tube as in an ordinary microscope. This helps stability by reducing weight at the extremities. Two 1/4-inch nuts are welded on the sides and near the ends of the vertical coupler of the welded pair (Fig. 5). The axis of the holes in the nuts should practically coincide to prevent binding when using. A long, threaded 1/4-inch bolt is screwed through both of these nuts far enough so that its lower end rests on the flange. A wing or knurled nut is welded to the bolt top for convenient finger turning.

The microscope may be swung in a circle after the winged bolt in the flanged coupler is tightened and the one in the vertical coupler is loosened. The microscope may be raised slowly by turning the long, threaded bolt clockwise. The lower end of this bolt thrusts against the rigidly held flanged coupler. The microscope may be slid in or out from the central axis or inclined at any angle from the vertical by loosening and tightening the winged stove bolt.

In use the apparatus is placed on a table beside potted wheat plants. The assembly may be moved up or down to the desired position and then held in position by tightening a thumb nut. Fine focusing then may be done with the long bolt.

FIELD CROPS RESEARCH BRANCH, AGRICULTURAL RESEARCH SERVICE, UNITED STATES DEPARTMENT OF AGRICULTURE, IN COOPERATION WITH THE DEPARTMENT OF BOTANY AND PLANT PATHOLOGY, KANSAS STATE COLLEGE, MANHATTAN, KANSAS.

VARIETAL RESPONSE OF CASTORBEAN TO TOBACCO RING SPOT VIRUS

A. A. Cook

Summary

Castorbean, Ricinus communis, was found to be susceptible to tobacco ring spot virus but not to nine other viruses with which it was inoculated. Tobacco ring spot virus was not transmitted with seed from infected plants. Three types of varietal responses to this virus were typified by the varieties Cimarron, Conner, and U. S. 49, in ascending order of severity. Histological examination of cotyledon petioles, hypocotyl, and epicotyl sections of inoculated Cimarron and U. S. 49 plants revealed only the epidermis of the epicotyl to be adversely affected in the former variety. In U. S. 49, the epidermis, adjoining parenchymatous tissues, phloem, and xylem, and related tissues were progressively more severely affected with increase in time after inoculation.

Castorbean, Ricinus communis L., was reported susceptible to infection with tobacco ring spot virus by Wingard (4) and by Holmes (3). Costa and Bennett (2) found castorbean not susceptible to a Euphorbia mosaic, but no other attempts at infection of this plant with a virus disease have been reported.

Preliminary inoculations of castorbean with the viruses of cucumber mosaic (two strains), tomato ring spot, tobacco ring spot, potato "Y" (three strains), tobacco etch (two strains), alfalfa mosaic, potato "X" (two strains), yellow bean mosaic, tomato aucuba, and tobacco mosaic (two strains) indicated only tobacco ring spot to be infectious. One plant inoculated with tomato ring spot virus exhibited slight symptoms but recovery tests were unsuccessful.

Further inoculations were undertaken to investigate possible differences in varietal reactions to tobacco ring spot, and a histological comparison was made between two varieties distinctly different in reaction.

Inoculation of Varieties

Seed of 32 varieties of castorbean were planted in 6-inch clay pots filled with methyl-bromide treated soil. Plants were thinned, if necessary, to five or six plants in each of the four pots planted to one variety in each experiment. Extracted juice from 14- to 21-day old systemically infected Cimarron castor bean or Nicotiana tabacum L. (Turkish) was used as inoculum. Test plants were dusted with 600-mesh carborundum when the cotyledonary leaves were fully expanded, and inoculated by the carborundum-gauze pad method. Plants were rinsed immediately after inoculation.

Record was made approximately every 4 days (starting 8 days after inoculation) of the symptom development on each plant, and continued through at least 36 days after inoculation.

Original inoculum was obtained by Dr. M. K. Corbett from Dr. A. F. Ross, Cornell University, and was maintained in Cimarron castorbean after the first inoculations.

All inoculations were completed between October 31, 1955 and March 2, 1956, so as to avoid, as nearly as possible, alteration of symptomology by change in air temperature.

Varietal Response to Tobacco Ring Spot Virus

Distinct differences in degree of susceptibility were apparent (Table 1). Infected plants of varieties Cimarron (Fig. 1) and N224A-1-1-1 always exhibited unmistakable evidence of infection, but only occasionally would one or more plants have a slight necrosis of the epicotyl. The disease was never fatal to a single plant of these two varieties. The variety Custer seemed to be largely composed of plants reacting in a similar manner to this disease except lacking the occasional necrosis of the epicotyl. However, approximately one of every four plants was killed by infection with this virus.

Varieties Baker 7, N145-4, N149-4-2, U. S. 49, U. S. 74, U. S. 121-6, and U. S. 243 were decidedly more severely affected by tobacco ring spot virus than the varieties mentioned above. As early as 8 days after inoculation, internal necrosis of the hypocotyl was macroscopically evident in some cases. The necrosis rapidly became more extensive in most instances until the plants were dead within 20 days after inoculation. With variety U. S. 49,

plants were frequently dead within 10 to 14 days after inoculation. Some individual plants of other varieties, however, would exhibit no foliar symptoms for as long as 20 days after inoculation, only subsequently to show hypocotyl necrosis (Fig. 2) and die. Some such plants were killed before foliar symptoms were apparent.

Table 1. Average disease index of castorbean varieties 36 days after inoculation with tobacco ring spot virus.

Variety	Source of inoculum: Tobacco[a]	Source of inoculum: Castor bean[b]	Variety	Source of inoculum: Tobacco	Source of inoculum: Castor bean
Anjou	-	84[c]	N294-2	41	75
Baker 1	-	80	U. S. 49	94	100
Baker 7	77	100	U. S. 74	79	96
Baker 34	-	92	U. S. 101	49	61
Baker 195	-	92	U. S. 121-6	-	100
Cimarron	26	30	U. S. 144	-	82
Conner	-	73	U. S. 162	-	91
French Organico	56	80	U. S. 243	-	98
Ill. 48-36	-	88	U. S. 250	-	90
Ill. 48-84	-	74	Custer	-	26
N145-4	66	98	W. O. 6	85	77
N149-4-2	79	100	W. O. 7	65	68
N224A-1-1-1	25	32	W. O. 10	-	84
N224A-1-6-4-3	65	87	U. S. 1420	-	55
N224A-2-1-1	64	85	Baker 158	-	65
N224A-2-1-3	-	60	Rall-15-2-B-B	-	65

[a]Tobacco inoculum used in two experiments with number of test plants of each variety ranging from 8 to 37, average 19.

[b]Castorbean inoculum used in 8 experiments with number of test plants of each variety ranging from 8 to 67, average 39.

[c]Infected plants were rated for severity of disease development from 25 - 100 where a rating of 25 = only foliar symptoms; 50 = necrosis of hypocotyl and/or petiole; 75 = extensive necrosis of hypocotyl; 100 = plant killed.

A third type of varietal reaction was noted wherein the full range of symptom expression could be found. This varietal reaction, considered intermediate between the two types described above, is typified by early "top necrosis" or killing of the growing point. It was not uncommon for all true leaves to drop with only the cotyledons remaining attached and green, and the axillary buds of the cotyledons or true leaves to begin growth. Axillary bud growth was always quite slow and malformed, although nearly all such plants continued to grow slowly until discarded. While this type of disease development was frequently encountered in the varieties classified herein, there were also found in each variety plants that were killed and plants that were much less severely affected. The more or less heterogeneous varieties, at least in regard to reaction to tobacco ring spot virus, that were classed in this intermediate category are Anjou, Baker 1, Baker 34, Baker 195, French Organico, Conner, Illinois 48-36, Illinois 48-84, N224A-1-6-4-3, N224A-2-1-1, N224A-2-1-3, N294-2, U. S. 101, U. S. 144, U. S. 162, U. S. 250, W. O. 6, W. O. 7, W. O. 10, U. S. 1420, Baker 158, and Rall-15-2-B-B.

For the first and third inoculations, inoculum was obtained from N. tabacum (Turkish), while inoculum for all other inoculations was obtained from Cimarron castorbean. Computation of a cumulative disease index for varieties that were inoculated with inoculum from both sources gave indications, in most cases, of less severe disease development with inoculum from tobacco than from castorbean (Fig. 3). This apparent variation in inoculum can not be attributed wholly to source plants with any degree of reliability, however, since no attempt was made to evaluate concentration of inocula. No evidence of seed transmission was obtained.

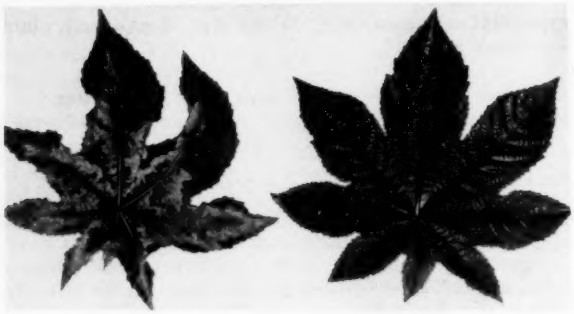

Fig. 1. Cimarron castorbean leaf systemically infected with tobacco ring spot virus (left) and healthy leaf (right).

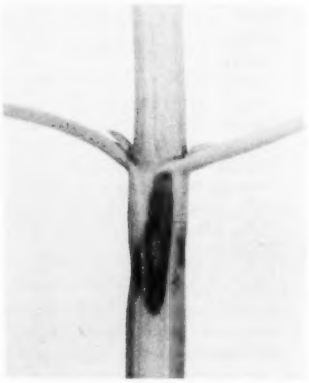

Fig. 2. Necrotic lesion on hypocotyl of N224-2-1-1 castorbean plant 25 days after mechanical inoculation with tobacco ring spot virus.

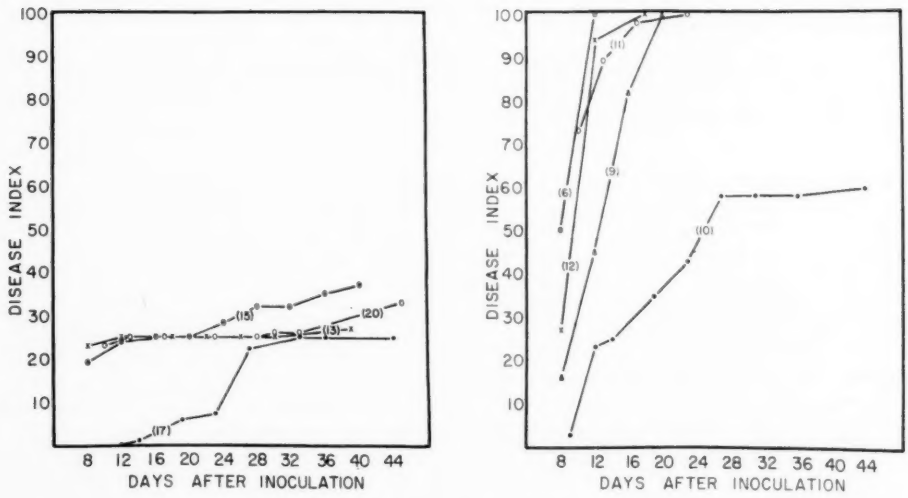

Fig. 3. Cumulative disease indices for 4 experiments using Cimarron (left) and 5 experiments using N149-4-2 (right) castorbean plants infected with tobacco ring spot. Inoculum from N. tabacum used in (.) and (0) experiments; from Cimarron castorbean for other experiments. Numbers in parentheses indicate number of infected plants in each respective experiment.

Histology

Varieties Cimarron and U. S. 49 were planted and inoculated 19 days later. At intervals of 2 days, for the succeeding 14 days, material was collected from five inoculated plants of each variety. Material was also collected from the same number of Cimarron plants at 20- and 28-day intervals after inoculation. From plants the same age as those inoculated, uninoculated plant material of both varieties was collected at intervals of 4, 8, and 12 days, and also from Cimarron 20 days from date of inoculation.

Specimens were taken from the petiole of cotyledons, the upper 25 mm of hypocotyl, and in some cases from the extreme lower 6 mm of epicotyl. Immediately after collecting, the pieces were killed in a formalin-acetic acid-alcohol solution and evacuated. Dehydration and infiltration was accomplished by a tertiary butyl alcohol series, and 52-52° C tissuemat was used for embedding. Sections were cut 12 microns in thickness and subjected to a quadruple stain procedure (1).

Local lesions were macroscopically evident on the cotyledons of U. S. 49 as early as 4 days after inoculation, while the first microscopic evidence of infection in hypocotyl and petiole sections was found 2 days later. Xylem parenchyma, xylem, and phloem became progressively more severely affected until plants of this variety were killed by 14 days after inoculation (Fig. 4). Extensive collapse of the epidermis and adjacent parenchymatous tissues was evident in the petioles at this time, the same symptoms being present but to a lesser degree throughout those portions of the hypocotyls examined.

Fig. 4. Cimarron (left) and U. S. 49 (right) castorbean plants 10 days after mechanical inoculation of cotyledons with tobacco ring spot virus.

The only internal symptom on Cimarron, which exhibited local lesions on the inoculated cotyledons 6 days after inoculation, was a necrosis that involved only particular areas of the epidermal layer of the epicotyl.

Discussion

While tobacco ring spot virus was not obtained from naturally infected castorbeans, no variety tested was found to be immune. However, if this disease should become of sufficient economic significance to warrant control by breeding new varieties, two reaction types could be utilized: 1) Cimarron-type reaction in which no hypocotyl necrosis accompanies foliar symptoms and infected plants "recover" to a certain extent; 2) hypersensitive reaction characterized by U. S. 49 in which all infected plants are killed.

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SUGAR BEET YELLOW VEIN DISEASE

C. W. Bennett[1]

Summary

A disease of sugar beet characterized by yellowing of veins of the leaves, dwarfing of one side of the plant in early stages of infection, and stunting of the entire plant in later stages of development, has been reported in Arizona, California, Colorado, Kansas, and New Mexico. Infected plants are damaged severely but the incidence of infection has been low, so far as reported. The causal agent is graft-transmissible in sugar beet and apparently is a virus. It was not transmitted by juice inoculation and no transmission was obtained with the common insects, such as the green peach aphid (Myzus persicae Sulz.), the black bean aphid (Aphis rumicis L.), the potato aphid (Macrosiphum solanii Ashm.), a green leafhopper (Empoasca sp.), and the beet leafhopper (Circulifer tenellus (Baker)), that feed on sugar beet. The virus apparently is not seed-transmitted and no transmission was obtained with three species of dodder -- Cuscuta californica Choisy, C. campestris Yuncker, or C. subinclusa Dur. & Hilg.

INTRODUCTION

A disease of sugar beet, Beta vulgaris L., has appeared annually in experimental plots and in commercial fields near Riverside, California, since 1936. The disease was found also near Lancaster, California, and specimens have been received at various times from Arizona, Colorado, Kansas, and New Mexico. In fields in California where the disease has been found, infection has been limited to a few plants per acre. There is some evidence that the disease may have been more prevalent in some fields in Colorado in certain seasons.

Probably the first mention of this disease in the literature is by Robbins (4) in 1921. In describing plants affected by a disease different from sugar beet mosaic, Robbins states, "The plants are distinguished by their asymmetry. One-half of the plant, including both top and root, is dwarfed; the other half is apparently normal. The affected leaves are small, thick, curled, twisted, and the mesophyll tissue bordering the veins of the leaf is pallid. The root shows eccentric growth, and seldom attains normal size."

Bodine (2) reported that "vein mosaic" was found in a number of fields of sugar beet in Larimer County, Colorado, in 1943. Bennett (1) partially described the disease in 1944 and reported transmission by graft. Maxson (3) states that he observed the disease as early as 1913. Apparently, therefore, it has been present in sugar beet fields in Western United States for many years but there is no evidence that it has increased appreciably in geographic range or in prevalence in the areas where it occurs.

Since yellowing of the larger veins of the leaves of affected plants is so characteristic of the disease and typical mottling rarely, or never, occurs under field conditions, it is suggested that "Sugar Beet Yellow Vein" be retained as the name of the disease.

ECONOMIC IMPORTANCE

The low incidence of sugar-beet yellow vein under most conditions has relegated it to a position of minor importance as a disease of sugar beet up to the present time. In California the disease has been relatively rare. Maxson (3) reported infection up to 1 percent in certain fields in 1913 and stated that occasionally it became sufficiently prevalent to cause measurable crop damage. In more recent years the disease has been found in Arizona, Colorado, Kansas, and New Mexico, but information as to the amount of infection and the damage produced is not available.

Although apparently this disease has not caused serious losses to the beet crop as a whole, it is capable of causing severe reduction in yield in infected plants. Maxson (3) reported that weight of roots of infected plants was reduced nearly 60 percent and that sucrose content was reduced also. Plants infected when in the earlier stages of growth are much reduced in size and widespread infection undoubtedly would result in serious losses.

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SYMPTOMS

The earliest symptoms of this disease begin to appear on very young leaves of infected plants and consist of yellowing of the main veins and dwarfing of the affected leaves. As the disease progresses, plants are stunted severely. Often the dwarfing is more marked on one side of the plant, especially during the first weeks following the appearance of symptoms (Fig. 1). Under greenhouse conditions dwarfing may or may not be more marked on one side of the plant, probably depending on the point of initial infection and on rate of lateral spread of the causal agent. In later stages, usually all of the new leaves show symptoms.

Fig. 1. Sugar-beet plant affected by yellow vein and showing asymmetrical development of the top and yellowing of veins of leaves on the affected side of the plant.

Fig. 2. Sugar-beet leaf from plant with yellow vein disease showing yellowing of tissue around the larger veins typical of symptoms produced by the disease.

Under field conditions the main veins of all affected leaves are distinctly yellow and the yellowing often extends into the adjacent parenchyma a distance of 1 mm or more (Fig. 2). The yellow areas may be continuous along the large veins or they may be broken. Frequently, on the smaller veins, there are more limited yellow spots isolated from other yellow areas, suggesting invasion of parenchyma tissue from local centers of spread. The yellow stripes and spots are visible on both sides of the leaf. Under greenhouse conditions, diffuse yellowing often is more pronounced. Yellowing, in some instances, is not closely limited to the larger veins and adjacent parenchyma and may include large areas of the leaf blade.

TRANSMISSION OF THE DISEASE

All available evidence on spread of the disease under natural conditions indicates that infection occurs early and probably comes from an outside source. No evidence has been found of spread from beet to beet in the field. Diseased plants have appeared as early as March 23 in seed fields near Phoenix, Arizona. Usually the disease appears later than this in southern California. Extensive inoculations have been made in an effort to transmit the disease under

experimental conditions.

Inoculation by Means of Juice: In 1939, 1942, and again in 1955 and 1956, attempts were made to transmit the causal agent of sugar-beet yellow vein by means of juice expressed from diseased plants. Several methods of preparing inoculum and numerous variations of the rubbing technique of inoculation were employed. More than 500 beet plants were inoculated but none developed symptoms of the disease.

In addition to sugar beet, plants of the following species were inoculated with juice from infected beets: Beta macrocarpa Guss., Brassica incana (L.) F. W. Schultz, Chenopodium album L., C. capitatum (L.) Asch., C. murale L., Cucumis melo L. (cantaloupe, var. Rocky Ford), Datura stramonium L. (jimson weed), Nicotiana acuminata Hook., N. glauca Graham (tree tobacco), N. glutinosa L., N. paniculata L., N. tabacum L. (tobacco, var. Turkish), and Phytolacca decandra L. (pokeweed). Twenty plants, or more, of each species were inoculated. None became infected.

Tests with Dodder: Three species of dodder -- Cuscuta californica Choisy, C. campestris Yuncker, and C. subinclusa Dur. & Hilg. -- were established on diseased sugar-beet plants. Strands of dodder were trained from diseased to healthy plants, and contact between the two types of plants was maintained for periods of about 7 days. Forty plants were inoculated with each species of dodder, and the inoculated plants were kept under observation for 2 months, or more. None of the inoculated plants became infected.

Transmission by Tissue Union: Several experiments were made in the transmission of the yellow-vein virus by tissue union. In one type of test, roots were inoculated by inserting slices of diseased tissue, about 5 mm thick, 2 to 3 cm long, and the width of the diseased crown from which they came, into appropriate slits made in the crowns of healthy roots. The diseased tissue was held firmly in place by cloth wrappings around the crown. Other plants were inoculated by inserting plugs of diseased tissue into the crown or root. All inoculated plants were incubated at about 45°F for a period of 20 to 30 days after which they were potted and placed in a greenhouse. There was a high percentage of infection in all tests, and transmission appeared to be obtained in all instances where tissue union occurred. The time for appearance of symptoms after transfer to the greenhouse varied from 16 to 30 days, in most instances, but in some plants, symptoms were delayed as long as 4 months.

Tests with Insects: Attempts were made to transmit the yellow-vein virus by means of the green peach aphid (Myzus persicae Sulz.), the black bean aphid (Aphis rumicis L.), the potato aphid (Macrosiphum solanii Ashm.), a green leafhopper (Empoasca sp.) from beet, and the beet leafhopper (Circulifer tenellus (Baker)). The species of aphids were colonized on diseased plants and transferred to healthy plants by placing infested, diseased leaves on the plants to be inoculated and allowing the aphids to crawl to the healthy leaves as the diseased leaves wilted. Large numbers of aphids were used on each plant. Sixty-four plants were inoculated with each species of aphid but none became infected.

The leafhoppers also were used in large numbers. After feeding 1 to 7 days on diseased plants they were transferred to healthy plants and allowed an additional feeding period of 7 to 14 days. Twenty plants were inoculated with each type of leafhopper. None showed indication of infection.

Tests with Seeds: In the spring of 1939, two plants affected by yellow vein flowered in the greenhouse and produced a fair crop of seed. The seeds were planted in flats and the seedlings were later placed in pots and kept until they developed 12, or more, true leaves. Of 481 seedlings tested, none developed symptoms of yellow vein. It seems likely, therefore, that the causal agent is not seed-transmissible.

CONCLUSIONS

Type of symptoms, pattern of spread, and graft transmissibility combine to indicate that the yellow-vein disease of sugar beet is caused by a virus. The disease appears to be distinct from any other disease of sugar beet. It most closely resembles yellow net described by Sylvester (5). However, yellow net causes less stunting and more extensive yellowing of the smaller veins, and yellowing does not extend so far into the parenchyma from the larger veins Also, yellow-net virus is readily transmissible by the green peach aphid, whereas yellow-vein

virus is not. Spread of yellow-vein virus probably is entirely by means of an insect vector, as yet unknown. Probably, however, sugar beet is either very resistant to infection or the vector is scarce or inefficient. It seems unlikely that the vector is an insect that feeds extensively on sugar beet since no evidence of spread from one sugar-beet plant to another has been observed. The source of inoculum is unknown, but the greater incidence of the disease in areas adjacent to buildings suggests that the virus may be associated in some manner with cultivated plants around the home, possibly some ornamental species, though no direct evidence has been obtained to support this hypothesis.

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FIELD CROPS RESEARCH BRANCH, AGRICULTURAL RESEARCH SERVICE, UNITED STATES DEPARTMENT OF AGRICULTURE, SALINAS, CALIFORNIA

STUDIES IN THE TREATMENT OF POTATO SEED PIECES WITH ANTIBIOTIC SUBSTANCES IN RELATION TO BACTERIAL AND FUNGOUS DECAY

Reiner Bonde[1] and Jean F. Malcolmson[2]

Abstract

Of the antibiotic substances tested, those containing streptomycin sulfate, streptomycin nitrate, and Terramycin hydrochloride gave good control of bacterial seed-piece decay. Material containing streptomycin sulfate was considered the most effective.

Nine thousand pounds or more of cut seed potatoes may be treated in 100 gallons of Agrimycin without reduction in the antibiotic activity of the solutions. Solutions containing streptomycin nitrate are less effective in similar treatments.

Solutions of Agrimycin maintain their antibiotic activity for at least three days after preparation.

Seed pieces treated with streptomycin solutions show an increased amount of Fusarium and Phoma rots and these fungi grow more rapidly on streptomycin-treated than untreated potato slices.

Introduction

In Maine the treatment of potato seed pieces with Agrimycin and streptomycin nitrate solutions has been found to be effective in controlling bacterial seed-piece decay and in reducing the amount of blackleg subsequently occurring in the field (1). These observations were confirmed in 1955 when different antibiotics were tested regarding their ability to control soft rot. Experiments were also conducted to determine the quantity of potatoes which may be treated effectively with solutions of Agrimycin 100 and Phytomycin, and to test the effect of storage on solutions of Agrimycin 100. Observations on the effect of Agrimycin 100 on fungous decay were also made.

Tests with Antibiotics for the Control of Soft Rot

The antibiotic substances listed in Table 1 were tested for their ability to control soft rot caused by Erwinia atroseptica, and observations were made on their effect on field stand.

Freshly cut potato seed pieces were dipped for 1 minute in a solution of the antibiotic under test, drained to remove excess moisture, and spray-inoculated with Erwinia atroseptica. The treated seed pieces were then stored in moisture-proof bags and examined for decay after 6 days. The treated seed pieces were planted in the field and their subsequent emergence was recorded. Inoculated control seed pieces to which no antibiotics were applied were treated similarly.

The data in Table 1 indicate that all the streptomycin solutions gave complete control of the seed-piece decay without reducing field emergence. Phytomycin gave complete control of the bacterial decay but in other experiments (Table 2) this substance was somewhat less effective than Agrimycin. Terramycin also gave good soft rot control in these experiments but in previous work (2 and 3) this has proved less effective than the different forms of streptomycin. PA-122, Magnamycin, and Malucidin did not have the ability to control bacterial soft rot.

Determination of the Quantity of Potatoes Which May Be Treated with Agrimycin 100 and Phytomycin

Experiments were conducted to determine the approximate quantity of potatoes that may be treated in solutions of Agrimycin 100 and Phytomycin without reducing the antibiotic activity of the solutions.

Forty-five 2-pound samples of freshly cut Kennebec seed pieces were dipped successively in 1-gallon solutions of the respective antibiotics at concentrations of 25, 50, and 100 ppm. Each dip simulated 200 pounds of seed potatoes in 100 gallons of solution on a field scale. The treated seed pieces were allowed to drain for 10 minutes, spray-inoculated with E. atroseptica, then incubated in moisture-proof, plastic bags. In cutting the seed pieces every sixth 2-pound

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Table 1. The effect of treatment with antibiotics on the control of bacterial seed-piece decay and field emergence.

Antibiotic[a]	Concentration (ppm)	Rotted seed pieces (%)	Field stand (%)
Soluble Terramycin Hydrochloride 1886-6-1	100	0.0	100.0
Terramycin Hydrochloride 1886-6-2	100	0.0	100.0
PA-122 1886-6-3	100	66.0	85.0
Acidulated Streptomycin Sulfate 1886-6-4	100	0.0	100.0
Magnamycin 1886-6-5	100	44.0	90.0
	50	36.0	90.0
Agrimycin 100	100	0.0	100.0
Streptomycin Sulfate	100	0.0	100.0
Agri-Strep Type A (Streptomycin Sulfate)	100	0.0	100.0
Malucidin (1% active)	100	60.0	40.0
	50	66.0	40.0
Phytomycin (Streptomycin Nitrate)	100	0.0	100.0
	50	0.0	100.0
	25	0.0	100.0
Control (untreated and inoculated)	—	80.0	35.0

[a] The first five antibiotics listed were supplied by Pfizer Company and the remainder by General Chemical Company, American Cyanamid Company, Merck Chemical Company, Naugatuck Chemical Company, and Olin Mathieson Company, respectively.

Table 2. Effect of repeated use of an antibiotic solution for the control of seed-piece decay caused by Erwinia atroseptica.

Antibiotic	Concentration (ppm)	Percentage rot in inoculated seed pieces after: Antibiotic treatment	No antibiotic treatment[a]
Agrimycin	25	0	88.6
	50	0	99.3
	100	0	79.2
Phytomycin	25	6.76[b]	72.7
	50	0.8[c]	84.2
	100	2.22[d]	86.9

[a] Each figure is the average rot in nine 2-pound control samples, expressed as a percentage of seed pieces infected.

[b] 80, 82, 95, 4, 39, and 4 percent seed pieces rotted in dips number 3, 4, 5, 12, 24, and 40 respectively.

[c] 12, 4, 4, 12, and 4 percent seed pieces rotted in dips number 9, 11, 14, 21, and 25 respectively.

[d] 16, 4, 4, 12, 4, 8, 8, 8, 16, 8, 8, and 4 percent seed pieces rotted in dips number 5, 14, 21, 22, 30, 32, 35, 36, 37, 39, 40, and 45, respectively.

FIGURE 1. Effect of treating seed pieces with 25 ppm Agrimycin 100. Left: Kennebec plants from inoculated seed pieces that had been treated with the Agrimycin solution after it had been used for 30 dip treatments. Right: No plants from inoculated seed pieces that had not been treated with Agrimycin.

sample was treated as a check in which the seed pieces were inoculated but received no antibiotic treatment.

The results of the experiment, given in Table 2, indicate that in all the treatments Agrimycin 100 gave complete control of bacterial decay. This indicates that 9,000 pounds (150 bushels) of cut seed potatoes can be treated in 100 gallons of Agrimycin solution, even at as low a concentration as 25 ppm, without the solution losing its antibiotic activity. Phytomycin was less effective and some rotting occurred at all concentrations.

Ten seed pieces from each treatment were planted in the field to determine the effect of the treatment on emergence. No injury due to the antibiotics was apparent and all the seed pieces produced healthy, vigorous plants. However, the control series, which contained a high percentage of decayed seed pieces, gave rise to weak plants or missing hills in the field (Figure 1).

The Effect of Storage on Agrimycin 100 Solutions

Experiments were set up to determine whether Agrimycin 100 solution would deteriorate if stored before use. A solution of 100 ppm Agrimycin 100 was prepared and used for treating one 2-pound seed lot on each of five successive days. The treated seed pieces were spray-inoculated with E. atroseptica and stored in moisture-proof plastic bags.

The treating solutions were found to give good control of bacterial decay for 3 days after preparation but after that their antibiotic activity was reduced.

The Effect of Treatment with Antibiotics on Fungous Decay

Although treatment with streptomycin has been effective in controlling bacterial soft rot in potato seed pieces it has not inhibited fungous decay (Figure 2). Indeed, among the treated seed pieces an increase in the rots caused by Fusarium and Phoma has been observed. This difference has been commonly attributed to suppression by the streptomycin of bacteria which normally inhibit fungous growth. To test this theory, an experiment was made to determine the effect of streptomycin sulfate on fungous pathogens under aseptic conditions.

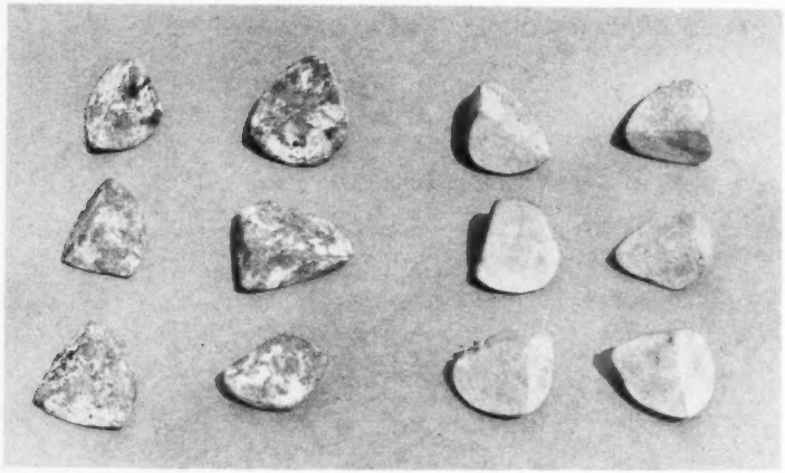

FIGURE 2. Effect of Agrimycin on mould growth on potato seed pieces. Left: Seed pieces treated with Agrimycin showing considerable mould growth on the surface. Right: Seed pieces not treated with Agrimycin showing no surface mould.

FIGURE 3. Effect of Agrimycin on the growth of Fusarium. Left: Untreated potato slices inoculated with Fusarium. Right: Agrimycin-treated potato slices, inoculated with Fusarium, showing increased Fusarium growth.

Slices 15 mm thick were cut from surface sterilized potatoes and dipped for 5 minutes in a 200 ppm Agrimycin 100 solution. The slices were then inoculated with two standard-sized discs of a common tuber-rotting Fusarium or of Phoma tuberosa and incubated under moist conditions in Petri dishes. A check series treated similarly, but in which the slices were dipped in sterile water in place of the Agrimycin solution, was also prepared.

After 3 weeks, growth of both fungi in depth and surface area was much greater in the Agrimycin-treated slices than in the check series (Figure 3). Treatment with Agrimycin, therefore, must either increase the susceptibility of the potatoes or stimulate the growth of these fungi, or both. Alteration of the biological equilibrium by the streptomycin is not necessarily responsible for the increased fungous decay under field conditions.

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MAINE AGRICULTURAL EXPERIMENT STATION

FOLIAGE FUNGICIDES PLUS GLYCERIN FOR THE CHEMOTHERAPY OF CUCUMBER SCAB

Saul Rich

Summary

Captan, zineb, and streptomycin with and without added glycerin were sprayed on cucumber seedlings at the two-leaf stage. After the third leaf had developed, the plants were inoculated with Cladosporium cucumerinum. The amount of disease developing on the third leaf, which had received no spray, was taken as an estimate of the chemotherapeutic activity of the treatments. Zineb and captan showed significant chemotherapeutic activity. The addition of glycerin significantly improved the chemotherapeutic activity of zineb. The chemotherapeutic activity of captan and streptomycin was improved non-significantly by the addition of glycerin.

Cucumber scab, caused by Cladosporium cucumerinum, is not adequately controlled in Connecticut by field spraying. One of the factors contributing to the field failure of fungicides is the difficulty of covering the undersides of the large, limpid, ground-hugging leaves of the mature cucumber vines. Fruit protection is also difficult because of the speed with which the fruits develop. Chemotherapeutants might protect these inadequately covered leaves and fruit.

Stoddard (5) has found that captan acts as a chemotherapeutant against cucumber scab. In spite of this activity by captan, it is still not adequate on cucumbers in the field. Zineb, chemotherapeutically active against blue mold of tobacco (6), controls scab on open-growing summer squash, but is not effective in the field against scab on cucumbers (1). The experiment reported here is an attempt to increase the chemotherapeutic activity of these two fungicides and streptomycin by the addition of glycerin (2). Streptomycin was tested for two reasons. First, it is translocatable when applied to above-ground plant parts (3). Second, it was found in preliminary experiments to be a weak protectant against cucumber scab.

The concentrations used were 2500 ppm of a 50% captan wettable powder, 2500 ppm of a 65% zineb wettable powder, 200 ppm streptomycin in a 15% wettable powder, and 10,000 ppm of glycerin.

Cucumbers, variety Marketer, were grown in sand, 5 plants per box. When the plants showed two true leaves, they were sprayed once, three boxes per treatment, and kept in the greenhouse at 80°F. When the third leaves were partially developed, 4 days later, the plants were inoculated with a spore suspension of C. cucumerinum. The spore suspension contained cucumber leaf sap to insure spore germination (4). The inoculated plants were incubated overnight in a moist chamber at 65°F. Afterwards they were kept in a greenhouse at 70°F until symptoms developed. To test the chemotherapeutic activity of the treatments, the amount of scab developing on the third leaf, which had received no spray, was estimated on the sixth day after inoculation. The disease readings were graded in severity from 0 for no disease, to 10 for complete death of the leaf. None of the treatments was phytotoxic.

The results are given in Table 1. The treatments in order of chemotherapeutic activity were captan plus glycerin, zineb plus glycerin, captan alone, zineb alone, and, finally, streptomycin plus glycerin. Streptomycin alone, and glycerin alone were grouped with the check as being entirely ineffective.

Treatments containing captan and zineb were significantly better than the check. Zineb was significantly improved by the addition of glycerin. Both captan and streptomycin, however, showed non-significant improvement by the addition of glycerin.

These results suggest that glycerin would improve the field performance of captan and zineb. The addition of glycerin would probably be worthwhile for controlling scab on summer squash. It is questionable whether or not glycerin would improve the field performance of these fungicides sufficiently to protect cucumbers against scab.

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Table 1. Chemotherapeutic activity of spray applications for the control of cucumber scab. Added glycerin used at 10,000 ppm.

Material	Concentration (ppm)	Average severity of disease on third (unsprayed) leaf[c]
Captan	2500 of formulation[a]	5.6
Captan plus glycerin	"	4.1
Zineb	2500 of formulation[b]	7.1
Zineb plus glycerin	"	4.9
Streptomycin	200	10.0
Streptomycin plus glycerin	200	8.2
Glycerin	10000	10.0
Check	—	9.7

[a] 50% wettable powder

[b] 65% wettable powder

[c] Severity rating: 0 for no disease, to 10 for complete death of leaf. L.S.D. at the 20% level = 1.9

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FIELD EXPERIMENTS FOR CONTROL OF BACTERIAL WILT OF SWEET CORN BY ANTIBIOTIC AND TWEEN 20 SPRAYS[1]

John L. Lockwood[2] and Lansing E. Williams

SUMMARY

Sprays containing 100 ppm streptomycin or 50 ppm Terramycin reduced the incidence and severity of bacterial wilt of sweet corn at two locations in Ohio. Yields, however, were not increased. Higher concentrations of the antibiotics gave greatest disease control. Tween 20, at the concentration used as the wetting agent (1000 ppm), decreased disease in the plots at one area, but differences were not significant at the other. Foliage sprays of 10,000 ppm, and "whorl" applications of 3000 ppm Tween 20 also reduced disease in some instances, but were severely phytotoxic.

INTRODUCTION

Antibiotic sprays have been effective in controlling several bacterial stem and leaf diseases, but few attempts have been made to control vascular wilt diseases with these materials. Williams and Lockwood (3) obtained some control of bacterial wilt of cucumber in the field with streptomycin and Terramycin sprays. Natti (1) reported that streptomycin sprays reduced the spread of bacterial wilt of sweet corn (Bacterium stewartii E. F. Sm.). In greenhouse tests prior to the field studies reported herein (4), the severity of bacterial wilt of sweet corn was decreased by Terramycin sprays of 50 ppm. The degree of control was enhanced by placing plants in a moist chamber for 24 hours before inoculation. Wilt symptoms were not reduced by sprays of 500 ppm streptomycin except when the moist chamber treatment was used. Tween 20, the wetting agent used, was effective in reducing the severity of bacterial wilt when 10,000 ppm solutions were applied as foliage sprays or 1,000 ppm solutions introduced into the "whorl". To test the possibility that antibiotics or Tween 20 might be effective in controlling bacterial wilt of sweet corn in the field, experiments were conducted at two locations in Ohio in 1955.

PROCEDURE

Experimental plots were located at Marietta and Wooster, Ohio. North Star, an early maturing hybrid sweet corn susceptible to bacterial wilt, was used as the test plant. Plots were in 4 randomized blocks consisting of 9 one-row treatments. Seed were planted in rows 3 feet apart with a drill which dropped approximately one seed per foot. Seedlings were not thinned.

Materials tested were the antibiotics streptomycin (Merck STS) and Terramycin (Pfizer soluble Terramycin), and the surface active agent Tween 20 (Atlas Powder Co.). The antibiotic spray solutions were carried in 1000 ppm Tween 20.

Materials were applied with a portable compressed air sprayer. Sprays were applied to the top, and, insofar as possible, to the bottom surfaces of the leaves. Whorl applications were made with a wash bottle. No insecticides were applied. Natural incidence of disease was sufficient at both locations, consequently no artificial inoculations were made.

Plots at Marietta were seeded on April 18. Rows in each block were 50 feet long. Approximately 200 plants received each treatment. All materials were applied May 2 (emergence), May 6, May 10, May 15, May 18, and May 25; on June 1 and June 8, 500 ppm Terramycin and 3000 and 10,000 ppm Tween 20 were omitted because of severe phytotoxicity.

Plots at Wooster were seeded on May 13. Rows in each block were 68 feet long. Approximately 280 plants received each treatment. All materials were applied on May 24 (2-leaf stage), May 27, and May 31; on June 3, June 9, and June 16 whorl applications of Tween 20 were omitted because of severe phytotoxicity.

Disease indices and yields were determined at each location. Each plant in the field was given a rating from 0 to 5. Zero indicated a healthy plant and 5 a dead plant. Criteria used in assigning ratings were degree of stunting, number of diseased leaves, and severity of leaf

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damage. Rating means calculated for each row served as disease indices. Disease was rated twice at each location, once when plants were about a foot high, and at maturity. All ears were harvested and wieghed at a date when most of the ears were mature.

Analyses of variance were made of disease indices, yield data, and disease incidences. As the number of plants per treatment for each block was less than 100, arc sine transformations were made of disease percentages (2).

RESULTS

Corn flea beetles (Chaetocnema pulicaria Melsheimer) were abundant at Wooster and Marietta at emergence of the corn and throughout the growing season. Although incidence of bacterial wilt was high at both locations in 1955, the disease was not so destructive as in 1953 and 1954 when many fields were ruined by the disease. Most diseased plants in 1955 grew and produced marketable ears.

The antibiotic sprays reduced bacterial wilt, as measured both by disease index and by disease incidence at both locations (Tables 1 and 2). There was marked reduction of disease in rows sprayed with the higher levels of these antibiotics.

Applications of 1000 ppm Tween 20 at the Wooster plots markedly reduced incidence and severity of wilt when compared with the untreated control. Although disease in rows sprayed with 1000 ppm Tween 20 did not differ significantly from the untreated control plots, a similar trend was evident at Marietta. At Wooster, with few exceptions, disease indices and incidences for the lower level antibiotic treatments did not differ from the Tween 20 control. A greater degree of control, however, was given by the higher concentrations of these antibiotics Consequently, part of the effectiveness of the antibiotic treatments may be attributed to this wetting agent. In most cases, foliar sprays of 10,000 ppm, and whorl applications of 3000 ppm Tween 20 also reduced bacterial wilt in the Marietta plots. These treatments were very phytotoxic. At Wooster, control with 3000 ppm Tween 20 was no better than that obtained with 1000 ppm Tween 20, and in some instances it was not as good. At Marietta, these treatments were applied for a longer period and a greater degree of disease control was obtained, but phytotoxicity was more severe.

Antibiotic or Tween 20 treatments reduced disease indices and incidences but yields were not significantly increased. Antibiotic sprays at the Marietta plots reduced yields as compared with the untreated control, but only in the case of the higher concentrations of Terramycin when comparison was made with the Tween 20 control. Yields at Wooster were reduced by 500 ppm streptomycin when compared with rows receiving 1000 ppm Tween 20.

All streptomycin and Terramycin sprays caused some chlorosis of young plants. At maturity these chlorotic effects were hardly noticeable and plants were not visibly stunted. The phytotoxicity of 500 ppm Terramycin was most severe and reduced the stand of seedlings by about 50 percent.

Plants did not appear to be affected by 1000 ppm Tween 20, but higher concentrations of this material were severely phytotoxic. Phytotoxic effects were similar whether Tween 20 was introduced into the whorl or sprayed over the entire plant. Affected plants never grew more than a foot or two in height. Excessive tillers, usually 6 or 8, were produced. Leaves were dull in color with venation distorted and somewhat netted. Proliferation of the upper midrib frequently occurred. Another formative effect was an increase in the number of pistillate terminal inflorescences. A count of all plants at the Marietta plots showed an average of 6 percent pistillate terminal inflorescences for all treatments, except on plants treated with 3000 and 10,000 ppm Tween 20. These 3 treatments produced plants with an average of 41 percent pistillate terminal inflorescences.

As corn smut, Ustilago maydis (DC.) Cda., was prevalent in the Marietta plots, a count of smutted plants was made. Percentage of diseased plants was calculated without regard to size or number of galls per plant. Analysis of the data showed that applications of 500 ppm Terramycin greatly reduced smut incidence (Table 1).

DISCUSSION

The mode of action of the antibiotics in reducing bacterial wilt of sweet corn is not understood. Natti (1) pointed out that several mechanisms may be operative: (a) systemic action of the antibiotic, (b) inactivation of wilt bacteria in flea beetle lesions before infection, and (c) inactivation of wilt bacteria on mouth parts or within the body of the flea beetle following ingestion of antibiotics with leaf tissues. The authors (4) found that bacterial wilt was reduced when plants were sprayed with Terramycin solutions of 50 ppm, then inoculated 24 hours later by

Table 1. Effect of foliar applications of streptomycin, Terramycin, or Tween 20 on bacterial wilt of sweet corn in the field (Marietta).

Treatment[a]	Conc., ppm	Disease index		Disease incidence, percent		Yield, lbs	Smut incidence, percent
		6/2	6/27	6/2	6/27		6/27
Streptomycin STS	100	1.3	2.3	31	64	74	24
Streptomycin STS	500	0.1	1.1	11	39	72	20
Sol. Terramycin	100	1.3	2.8	28	63	61	24
Sol. Terramycin	500	0.2	2.8	11	50	49	5
Tween 20	10,000	1.3	2.4	33	60	--	19
Tween 20[b]	3000	0.9	1.7	33	55	--	24
Tween 20[b]	10,000	0.7	1.5	29	49	--	20
Tween 20 control	1000	2.5	2.8	39	66	91	21
No treatment	----	3.0	3.2	41	72	114	17
LSD 5%		0.6	1.2	8	12	34	5
LSD 1%		0.8	1.6	11	17	47	6

[a]Streptomycin STS, containing 54% streptomycin base, supplied by Merck and Co., Inc. Soluble Terramycin, containing 21% Terramycin base, supplied by Chas. Pfizer and Co., Inc. Tween 20 supplied by Atlas Powder Co., Inc. Concentrations based on active material.

[b]Solution introduced into whorls only.

hypodermic injection at the base of the culm. Streptomycin was ineffective at these concentrations. When plants were placed in a moist chamber for 24 hours following application of the sprays, streptomycin at 500 ppm reduced disease, and control by Terramycin was enhanced. These results indicate that systemic action of the antibiotics may be involved in the reduction of wilt symptoms. It would seem reasonable that the antibiotics on leaf surfaces might also act as a toxic barrier to the entry of bacteria into the tissues of the host. Bacteria on mouth parts or in intestinal tracts may be inactivated when the carrier beetles feed on the antibiotic-covered foliage.

Tween 20 appeared to act systemically in lessening severity of wilt symptoms. In the greenhouse, 10,000 ppm sprayed on the foliage, or 1000 ppm introduced into the whorl 24 hours before inoculation, consistently reduced the severity of symptoms of bacterial wilt (4). In laboratory tests Tween 20 at a concentration of 100,000 ppm (10%) was non-toxic to B. stewartii on agar (4). The material thus appeared to modify the resistance of the host in some manner.

Results with Tween 20 in field tests were disappointing. Severe phytotoxicity resulted when the material was injected into the whorl at concentrations of 3000 ppm, and sprayed on the entire foliage at 10,000 ppm. In the Wooster experiment, 1000 ppm Tween 20 sprays reduced disease severity and were not injurious. It is doubtful that the degree of control obtained by 1000 ppm Tween 20 was sufficient to encourage further testing. Lack of disease reduction with this concentration in greenhouse tests was probably due to the fact that only single applications were made.

The use of streptomycin or Terramycin sprays to control bacterial wilt of sweet corn does not appear promising. Phytotoxicity was noted, and despite the fact that disease indices and incidences were reduced yields were not increased. It is possible that adjustments of formu-

Table 2. Effect of foliar applications of streptomycin, Terramycin, or Tween 20 on bacterial wilt of sweet corn in the field (Wooster).

Treatment	Conc., ppm	Disease index 6/28	Disease index 7/14	Disease incidence, percent 6/28	Disease incidence, percent 7/14	Yield, lbs
Streptomycin STS	100	1.3	1.9	30	33	162
Streptomycin STS	500	0.5	0.8	19	19	139
Sol. Terramycin	50	1.0	1.5	25	31	147
Sol. Terramycin	200	0.5	0.7	20	23	151
Tween 20	10,000	1.0	1.3	34	34	69
Tween 20[a]	3000	1.8	2.7	35	41	143
Tween 20[a]	10,000	1.6	2.5	34	41	147
Tween 20 control	1000	1.5	1.7	30	33	174
No treatment	----	2.5	2.7	39	40	162
LSD 5%		0.5	0.7	6	6	30
LSD 1%		0.7	1.0	8	8	41

[a]Solution introduced into whorls only

lation, schedule, and method of application might enhance control and reduce phytotoxicity. It is also possible that the beneficial effects might be more striking in a season when bacterial wilt is more devastating than it was in 1955, a comparatively mild year. The answers to such questions await further testing. This report establishes that bacterial wilt can be significantly reduced by application of antibiotic sprays.

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PRELIMINARY COMPARISON OF AGRI-MYCIN 100 AND COPPER COMPOUNDS FOR THE CONTROL OF WALNUT BLIGHT IN OREGON[1, 2]

P. W. Miller

In recent years, antibiotics have received considerable attention from plant pathologists as offering a new approach to the control of many plant diseases hitherto difficult to control. The literature on antibiotics in relation to plant disease control was recently reviewed by Zaumeyer (7).

Antibiotic formulations containing streptomycin have been used successfully in California for the control of walnut blight caused by Xanthomonas juglandis. In field tests carried on in that State, Ark (1, 2, 3, 4) and Ark and Charles (6) reported evidence of control of this disease by timely spraying with streptomycin sulfate at concentrations of 10 to 100 ppm. The best control was obtained with concentrations of 25 to 100 ppm. In 1956, Ark and Alcorn (5) reported that 500-ppm and 1000-ppm streptomycin-pyrophyllite dusts, respectively, gave good control of walnut blight and were somewhat better than a 10% Copper A Compound dust.

Investigations on the control of walnut blight by spraying with antibiotics were begun in Oregon in 1954. A preliminary report of the results of field tests carried on in 1954 and 1955 follows.

Materials and Methods: Agri-mycin 100 -- a powder formulation containing 15% streptomycin sulfate and 1.5% Terramycin -- was used exclusively in these tests.

In 1954, Agri-mycin 100 at the rate of 100 ppm was compared with Yellow Cuprocide (2 pounds to 100 gallons) in 50-tree plots in a Franquette walnut orchard at Scholls, Oregon. In 1955, the same orchard and plot sizes were used as in 1954, and Agri-mycin 100 at the rate of 25 ppm was tested comparatively with Copper A Compound (4 pounds to 100 gallons). All spray materials were applied with a speed (air-blast) sprayer. Three applications (in the early prebloom, late prebloom, and early postbloom stages of pistillate flower development) of each spray material were made per season.

Results: The results of tests in 1954 and 1955 are presented in Table 1. In 1954, Agri-mycin 100 at the rate of 100 ppm was just as effective as Yellow Curpocide 2-100 for the control of walnut blight. In 1955, Agri-mycin 100 at the rate of 25 ppm gave practically as good results as Copper A Compound 4-100. However, the 1955 tests are not considered conclusive as there was relatively little disease (4.9 percent) in the untreated plots. While Agri-mycin 100 at the rate of 25 ppm appears to be effective under mild disease conditions, it remains to be determined whether it will be effective at this concentration under epidemic conditions.

Table 1. A comparison of Agri-mycin and some copper spray materials for the control of walnut blight in western Oregon in 1954 and 1955.

Year	Material and concentration	Number of applications	Relative amount of foliage injury	Nuts examined (number)	Nuts infected (percent)
1954	Yellow Cuprocide 2-100	3	Moderate	1500	1.9
	Agri-mycin 100 ppm	3	None	1517	1.1
	Untreated	0	None	1000	11.6
1955	Agri-mycin 25 ppm	3	None	1500	0.6
	Copper A Compound 4-100	3	Trace	1500	0.9
	Untreated	0	None	2000	4.9

[1]Approved for publication by the Director of the Oregon Agricultural Experiment Station and by the Chief of the Horticultural Crops Research Branch, Agricultural Research Service, United States Department of Agriculture. Contribution of the Department of Botany and Plant Pathology of the Oregon Agricultural Experiment Station and the Horticultural Crops Research Branch, Agricultural Research Service, United States Department of Agriculture.

[2]Acknowledgment is made to Chas. Pfizer and Company, Inc., for donating Agri-mycin 100 for trial.

Agri-mycin 100 at the rate of either 25 or 100 ppm caused no foliage injury whereas both copper materials used caused a trace to a moderate amount of injury.

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HORTICULTURAL CROPS RESEARCH BRANCH, AGRICULTURAL RESEARCH SERVICE, UNITED STATES DEPARTMENT OF AGRICULTURE, CORVALLIS, OREGON

EVALUATION OF 2, 3, 5-TRIPHENYL TETRAZOLIUM CHLORIDE FOR OBTAINING PATHOGENIC TYPES FROM STOCK CULTURES OF HALO BLIGHT AND COMMON BLIGHT ORGANISMS

B. C. Smale and J. F. Worley[1]

To facilitate basic studies of the effects of antibiotics on plant diseases, we required highly pathogenic types of some bacterial pathogens. The color reaction of tetrazolium when associated with certain bacteria (1) has been of value to us in obtaining highly pathogenic types from stock cultures of Pseudomonas phaseolicola (Burkholder) Dowson, the causal organism of halo blight of beans. Tetrazolium was not useful, however, in obtaining highly pathogenic types of the common blight organism, Xanthomonas phaseoli (E. F. Sm.)Dowson.

Serial dilutions of 12-month-old stock cultures of P. phaseolicola were streaked on potato-dextrose agar containing sufficient tetrazolium to give a final concentration of 0.005% of the salt. Sterile tetrazolium was added aseptically to the melted agar just prior to pouring plates. The plates were incubated at 28°C and observed macroscopically at 24 hour-intervals for color differences between individual colonies.

Colonies of comparable size which varied in color from entirely red, to various proportions of red and white, to entirely white, developed. The color of the individual colonies was found to be related to pathogenicity. The red colonies were highly pathogenic, while the white ones were weakly pathogenic. All others were moderately or weakly pathogenic, the degree of pathogenicity depending upon the proportion of red and white color in individual colonies.

In these pathogenicity tests, individual colonies of comparable size and exhibiting different color reactions were tested on 6-day-old Black Valentine bean plants with primary leaves only partially expanded and still wrinkled. Inoculm was prepared by incubating each colony type, selected on the basis of color, separately in beef-broth medium. After 24 hours at 28°C the cultures were diluted with 3 volumes of tap water. The upper surfaces of primary leaves were dusted with carborundum and inoculated by rubbing with cheesecloth saturated with the diluted bacterial suspension. The leaves were immediately syringed with tap water to prevent injury by the medium.

The correlation of pathogenicity and red color of individual colonies of this bacterium is interesting in view of the results obtained by Kelman.[2] He reported for Pseudomonas solanacearum E. F. Sm. that fluidal white colonies with pink centers were highly pathogenic, while butyrous red ones were weakly pathogenic or non-pathogenic.

In experiments similar to those using P. phaseolicola, individual colonies from stock cultures of X. phaseoli displayed color differences, but the degree of pathogenicity exhibited different colored colonies was the same.

Finally, both common and halo blight organisms were freshly isolated from infected plants for study of the color reactions when they were cultured on tetrazoliun agar, as previously described. Isolation was accomplished by carefully peeling off a portion of the lower epidermis of an infected leaf and thus removing the contaminated surface. An exposed lesion was then touched with a sterile glass needle. A needle of smaller dimension than the lesion was found to be most satisfactory. The bacteria were transferred from the needle to a drop of sterile water on an agar plate and then streaked with a conventional platinum loop.

Newly isolated organisms of both species were highly pathogenic. The freshly isolated bacteria of X. phaseoli causes severe infection within 7 days as compared with 14 or more days when bacteria from the original stock culture were used.

On tetrazolium agar the recently isolated cultures of both organisms produced within 48 hours many entirely red colonies and practically no white colonies. The red colonies appeared to develop from individual bacteria since they were widely separated on the plates when the new isolation technique was used. In contrast, in the same period the stock culture produced red, red and white, and white colonies.

HORTICULTURAL CROPS RESEARCH BRANCH, AGRICULTURAL RESEARCH SERVICE

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[1] Physiologist and Biological Science Aid, respectively, Horticultural Crops Research Branch, Agricultural Research Service, United States Department of Agriculture, Beltsville, Maryland.

[2] Kelman, Arthur. 1954. The relationship of pathogenicity in Pseudomonas solanacearum to colony appearance on tetrazolium medium. Phytopathology 44:693-695.

PECTIC ENZYMES OF PHYTOPHTHORA PARASITICA VAR. NICOTIANAE, THE CAUSE OF BLACK SHANK OF TOBACCO

Akhtar Husain and Arthur Kelman[1]

Summary

Liquid culture filtrates of Phytophthora parasitica (Dastur) var. nicotianae (Breda de Haan) Tucker were tested for the presence of pectic enzymes. No pectin-methyl-esterase was detected. Culture filtrates contained a polygalacturonase that caused a rapid loss in viscosity of a 1.2% sodium polypectate solution. Chemical assay of the enzyme revealed that reducing groups formed in small amounts only after prolonged incubation. Deposits of pectic substances in vessels of diseased tobacco plants may result from the action of this fungus polygalacturonase.

Introduction

Black shank of tobacco caused by Phytophthora parasitica (Dastur) var. nicotianae (Breda de Haan) Tucker is characterized by a sudden wilting of the plants associated with the appearance of black lesions girdling the basal portion of the stem. Wilting of infected plants has been attributed to the effect of a systemic toxin formed by the pathogen (9, 10). However, studies by Powers (6) indicated that wilting is caused by local obstruction of water movement at the basal lesion. Powers associated cessation of water movement with the formation of tyloses and gums in the vessels. Schramm and Wolf (7) also found that xylem tissue is occluded and water movement is impeded in tobacco plants infected by the black shank fungus. In other vascular diseases where wilting occurs as a result of plugging of vessels, data have been obtained indicating that pectic enzymes are involved in the mechanism of wilting (1, 5, 7). As an aid in explaining the mechanism of wilt induction by the black shank fungus, this investigation was initiated to determine whether pectic enzymes are produced by the fungus in vitro.

Materials and Methods

Enzyme production was tested in the following media: potato dextrose broth, oat meal broth, malt extract broth, a synthetic medium used by Mehrotra (4), and a starch-glutamic acid medium (10). Moderate to good growth developed in the starch-glutamic acid medium; the organism did not grow well in any of the other media tested. Aeration of liquid cultures by shaking was tried but it proved to be unfavorable for rapid growth of the fungus.

For preparation of extracts for enzyme assay two highly pathogenic strains of P. parasitica var. nicotianae[2] (one from burley and one from flue-cured tobacco) were grown in 250 ml. flasks containing 50 ml. of the starch-glutamic acid medium (in still culture) at 28°to30°C. After 2 weeks growth, the medium was filtered through a Buchner funnel and the crude culture filtrate was used as a source of enzyme. When stored, the filtrate was covered with a layer of toluene and kept in the refrigerator at 5°C.

Pectin-methyl-esterase Assay: P.M.E. assay was carried out by measuring the amount of carboxyl groups formed following Kertesz's continuous titration method (3) and using 1.0% citrus pectin as substrate.[3]

Polygalacturonase Assay: P.G. activity was measured by 1) estimating the loss in viscosity of a sodium polypectate solution and 2) determination of reducing groups formed. Viscosity change was determined in size 300 Fenske-Ostwald viscometers at 30°C. (±0.1) using as a substrate 1.2% sodium polypectate solution buffered with citrate buffer at pH 5.0. Reducing groups were estimated by the Willsttäter-Schudel hypoiodite method as modified by Jansen and MacDonnell (2).

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[1] Research Assistant and Associate Professor of Plant Pathology, respectively. North Carolina State College, Raleigh, North Carolina.

[2] Isolates obtained from Dr. J. L. Apple, North Carolina State College, Raleigh, North Carolina.

[3] Citrus pectin and sodium polypectate were obtained from the California Fruit Exchange.

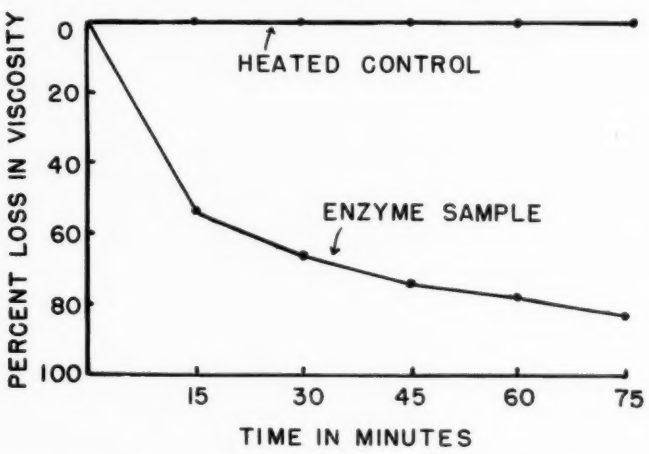

FIGURE 1. Loss in viscosity of a 1.2% sodium polypectate solution caused by polygalacturonase of Phytophthora parasitica var. nicotianae.

Results

The culture filtrate gave a negative test for pectin-methyl-esterase. However, it contained an enzyme which rapidly decreased the viscosity of a sodium polypectate solution. Figure 1 shows the percentage loss in viscosity at various time intervals by the action of this enzyme in a typical experiment. Similar results were obtained in three additional tests. When reducing groups were measured, no activity was detected in the first 5 hours. After incubation for more than 10 hours only small amounts of reducing groups were detected. Although the burley tobacco isolate grew better in the starch-glutamic acid medium than the flue-cured tobacco isolate, P. G. activity was slightly higher in filtrates of the latter.

Discussion

Wilting of tobacco plants affected by black shank can be attributed to the formation of tyloses and gums in vessels of the host (6). Powers considered that the formation of gums and tyloses was induced by the action of toxic substances produced either by the pathogen or the interaction of host and the fungus. The present findings provide a possible explanation for the formation of gums. The polygalacturonase produced by the fungus, by its action on the middle lamellae of cells and pit membranes of vessels, may produce fragments of pectic materials. When released into the vessels, these pectic materials may interact with other substances resulting in formation of gums. Thus, water movement through the vessels can be hindered. The fact that the gums present in the vessels of diseased tissue consist mostly of pectic substances supports this hypothesis (6). Furthermore, in Fusarium wilt of tomato it has been shown that pectic enzymes of the pathogen as well as commercial pectic enzymes are capable of producing viscous deposits of pectic materials in the vessels (5) similar to those observed by Powers in xylem tissue of diseased tobacco plants. Softening of the pit membranes of the vessels by the enzymes may also be involved in tylose formation.

In addition to pectic enzymes the moderately-heat-stable, dialyzable, non-proteinaceous toxin studied by Wolf and Wolf (10) may be involved in the wilting mechanism. However, the exact role of this toxin in pathogenesis has not been elucidated.

In the investigation of this toxic principle by Wolf and Wolf maximum yields were obtained only after 4 to 6 weeks' growth of the black shank fungus in the synthetic starch-glutamic acid medium. Two week-old culture filtrates of the same medium were used in the present study on enzyme production. It is possible that the exzyme polygalacturonase may have been present

only at very low levels in 4-6 week-old cultures since inactivation of pectic enzymes in liquid media may occur with increasing age of the culture.

Further studies in vivo are needed before definite conclusions can be drawn with respect to the specific action of this enzyme in the living plant. In addition more information on the nutritional requirements of the black shank fungus will be necessary to obtain the optimal medium for growth that is requisite for critical investigations of fungus physiology.

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A CASE HISTORY OF THE PERSISTENCE OF HETERODERA ROSTOCHIENSIS WOLLENWEBER IN A TREATED FIELD

J. F. Spears[1], W. F. Mai[2], and D. O. Betz[3]

Data obtained by members of the Section of Nematology, Horticultural Crops Research Branch, from field experiments conducted during 1944-45, indicated the practicability of accomplishing a high degree of control of the golden nematode, Heterodera rostochiensis Wollenweber, with D-D (dichloropropene-dichloropropane) applied with shank applicator at a rate of 450 pounds per acre (1).

In 1946 Plant Pest Control Branch, Agricultural Research Service, applied D-D to 1556 acres of land infested with the golden nematode in Nassau County, Long Island, New York. The work was accomplished between July 27 and September 11 of that year. The fumigant was applied in a single application in furrows 12 inches apart at a depth of 6 inches. The land was rolled immediately after treatment.

Since infested soil contains many old empty cysts, the actual cyst count would be the same before and after treatment. Therefore, the method used for determining the effectiveness of the fumigant was to compare the percentage of cysts with viable contents present before and after treatment.

Viability determinations of cysts recovered from one of these properties designated as 46-T-3, containing 55 acres, has been conducted annually since 1946.

The crop history of the field is as follows: potatoes, 1944; grain, 1945; fallow, 1946; grain, 1947-48; natural flora, 1949-51; corn, 1952-54; and natural flora, 1955. Although frequent examinations were made, no volunteer potato plants or other known hosts were observed in the field after the D-D treatment.

Immediately prior to fumigation, soil samples were collected from each of three sections of the field. The samples consisted of about one tablespoon of soil taken to a depth of 1 to 2 1/2 inches with a trowel every 25 paces on a grid pattern. The entire sample was washed by a flotation and screening process. Cysts, selected at random from the screened material, were dissected under 90x magnification. Viability was judged on larval characteristics visible immediately following dissection of the cysts (2). Soil samples were obtained from the field and the percentage of viable cysts was determined just prior to fumigation, 17 days following fumigation, and yearly thereafter (Table 1).

Because of the higher viability obtained in 1951, a comparison was made between the viability obtained with the method previously employed and that obtained by exposure of individual cysts to potato root leachings. In 1951 soil was collected directly from the field treated with D-D in 1946. Soil moved in the fall of 1951 to isolated boarded plots at the Golden Nematode Research Laboratory was used for the 1952, 1953, 1954 tests. In obtaining this soil from the treated field to put in the boarded plots, a sampling procedure similar to the one previously described for collecting soil samples was employed. In this case, however, because of the large quantity of soil desired, a shovel was used for sampling instead of a trowel and the soil was transported in a steam-cleaned truck body. No crops have been grown since in these boarded plots.

Cysts from the boarded plots used for determining viability were washed from soil recovered to a depth of from 0-8 inches from these plots by a soil auger. For the hatching tests, individual cysts, selected at random, were placed in leachings in 2-cm Syracuse dishes. After exposure to leachings for 3 weeks, the emerged larvae in each dish were counted.

There was a high correlation between the number of cysts containing viable larvae when viability was determined by exposure to leachings and when cysts were opened and the enclosed larvae examined (Table 2). These data indicate that the higher viability reading obtained in 1951 was not due to the method employed for making viability determinations. No satisfactory explanation can be given to explain this increase.

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TABLE-1

PERCENT VIABLE CYSTS PRIOR TO AND FOLLOWING FUMIGATION WITH D. D

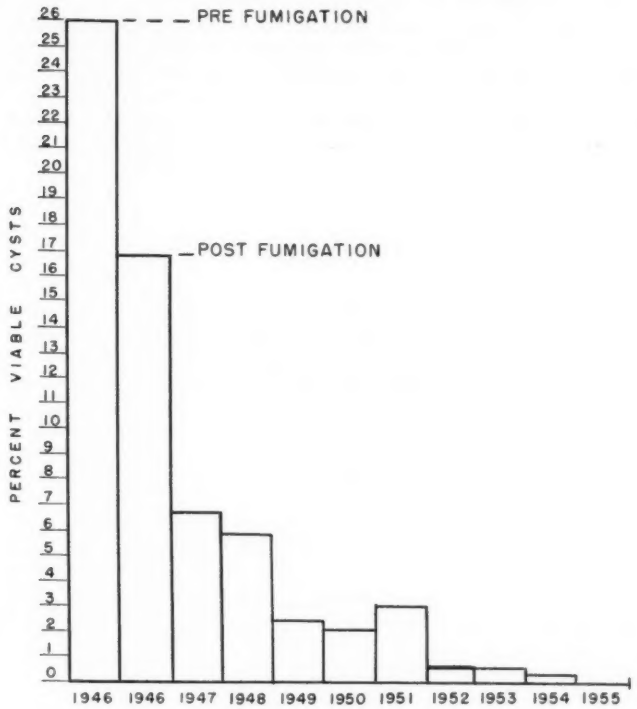

TABLE 2

DETERMINATION OF VIABILITY OF ENCYSTED LARVAE BY EXAMINATION AND BY EXPOSURE TO POTATO ROOT LEACHINGS.....

Viability Determined By				
	Examination		Exposure To Leachings	
Year	No. Cysts Examined	Percent With Viable Contents	No. Cysts Exposed To Leachings	Percent With Emerging Larvae
1951	1988	3.0	500	3.0
1952	400	0.5	400	0.5
1953	316	0.0	348	0.3
1954	296	0.3	343	0.0

In 1954, to check cyst viability further, a potato seed piece was planted in each of 24 4-inch clay pots filled with soil from the boarded plots. No golden nematode females were found on the roots in these pots. No larvae emerged from cysts exposed to potato leachings in 1954 and no cysts with viable larvae were found by the microscopic examinations made in 1955.

This record of a field which has been out of potato production for 9 years following soil treatment constitutes a case history of the combined effects of chemical and natural phenomena on the persistence of golden nematode encysted larvae in the soil under field conditions. The case history would be more complete if tests had been conducted each year to determine the relative numbers of females developing on potato roots. The ultimate test of viability is the capability to develop to maturity on host roots. At present, sufficient data are not available to establish the reliability of the results of visual examination and potato root leachings methods in predicting capability to enter and develop in potato roots.

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RESISTANCE TO THE STUNT NEMATODE IN CORN[1]

R. R. Nelson[2]

Abstract

Corn hybrids, inbreds, and single crosses were tested for their reaction to the stunt nematodes. The evaluation of lines of corn was based on a comparison of root weights and on nematode population studies. Inbred lines differed in their reaction to the stunt nematode. Susceptible inbreds had greatly reduced root systems and the population of nematodes increased under these inbreds. The number of nematodes decreased under resistant inbreds.

INTRODUCTION

A preliminary survey throughout the Coastal Plain of North Carolina in 1955 indicated that high populations of the tobacco stunt nematode (Tylenchorhynchus claytoni Steiner) were present in problem fields in corn in that area (2). Greenhouse studies demonstrated that the stunt nematode can cause severe damage to young corn plants and exhibits a marked population increase on that host. These findings pointed out the potential importance of the parasitism of corn by the stunt nematode, especially on corn that is grown in rotation with tobacco, a crop highly susceptible to the stunt nematode (1). Studies were then undertaken to determine whether resistance to the stunt nematode was present in commercial corn hybrids grown in the Coastal Plain and in the inbreds that comprise those hybrids.

MATERIALS AND METHODS

The initial evaluation of lines of corn was based on a comparison of total root weights of plants grown in sterilized soil with those grown in soil infested with the stunt nematode. Six inch pots were filled 1/3 full with steam sterilized soil and nematode infested soil then was added and covered with approximately 1 inch of sterilized soil. Five seeds per pot were planted and the young corn roots soon penetrated the nematode infested soil. Plant and root weights were recorded approximately 45 days after planting, which allowed sufficient time for one complete reproductive cycle of the stunt nematode on corn. At least two pots of each entry were tested. All of the hybrids commonly grown in the Coastal Plain of North Carolina were tested for their reaction to the stunt nematode.

RESULTS

When subjected to high populations of the nematode, plants of all hybrids had greatly reduced root systems, characterized by the lack of secondary or feeder roots (Fig. 1) and a marked reduction in vigor evidenced by reduced top growth. Primary roots frequently were discolored. The root weights of hybrids grown in infested soil were from 40 percent to 60 percent less than those of check plants. Similar results were obtained when the test was repeated. A single population study showed that the number of nematodes increased greatly when in contact with plants of all hybrids. These results indicated that none of the commercial hybrids tested were resistant to the stunt nematode.

Thirty inbred lines and 11 single crosses also were tested for their reaction to the stunt nematode. Twenty-one inbreds and all 11 single crosses showed a marked decrease in root weight and plant weight when exposed to high populations of the nematode. The most striking symptom was a reduction in the number of secondary roots (Fig. 2). Nine of the inbreds, which appeared to have some resistance to the stunt nematode, had weights of tops and roots similar to those of the check plants. There appeared to be only a slight reduction in number of secondary roots (Fig. 3) and primary roots seldom showed browning.

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[1] Cooperative investigations of the Field Crops Research Branch, Agricultural Research Service, United States Department of Agriculture and the North Carolina Agricultural Experiment Station.

[2] Plant Pathologist, Field Crops Research Branch, Agricultural Research Service, United States Department of Agriculture and Assistant Professor of Plant Pathology, North Carolina State College.

Table 1: Effect of 3 population levels of the stunt nematode on root weights of inbred lines and single crosses of corn.

Line	Initial infestation level - number of nematodes per pot								
	50			200			500		
	Aver. Root Wt.-Grams Ck.	Inoc.	Nematode [a] Population	Aver. Root Wt.-Grams Ck.	Inoc.	Nematode Population	Aver. Root Wt.-Grams Ck.	Inoc.	Nematode Population
Group A.									
TX 61M	5.0	4.7	+	7.7	5.3	+	13.5	4.9	+
CI 2A	6.1	6.2	+	9.0	6.2	+	17.1	10.1	+
L 10	7.5	7.2	+	7.6	4.4	+	18.1	6.2	+
T 204	7.7	8.2	+	8.5	5.1	+	8.8	3.5	+
F 44	9.1	9.0	+	14.2	9.7	+	13.4	7.6	+
Group A. Aver.	7.1	7.1 (100% of Check)		9.4	6.1 (65% of Check)		14.2	6.5 (46% of Check)	
Group B.									
NC 7	9.9	11.0	-	14.8	12.9	-	12.2	10.7	-
NC 89	5.8	5.1	-	6.4	5.3	-	6.5	5.4	-
NC 18	11.2	10.3	-	10.8	10.4	-	11.9	10.6	-
CI 14	9.0	8.7	-	10.1	8.4	-	9.7	9.2	-
T 7	9.5	9.3	-	11.2	10.4	-	11.4	12.7	-
CI 21	9.1	8.4	-	8.1	7.7	-	8.5	9.1	-
F 6	7.9	8.4	-	10.0	8.0	-	10.6	11.5	-
NC 16	8.5	7.2	-	5.6	4.7	-	13.6	11.7	-
NC 88	9.4	8.2	-	9.1	7.8	-	8.1	7.1	-
Group B. Aver.	8.9	8.5 (95% of Check)		9.5	8.4 (88% of Check)		10.3	9.7 (94% of Check)	
Group C.									
F6 X F44	15.2	13.2	+	15.0	11.4	+	16.1	11.0	+
NC 7 X T204	14.3	12.9	+	15.3	10.2	+	22.0	14.1	+
TX 61M X L10	10.3	9.1	+	11.6	8.2	+	14.2	7.1	+
Group C. Aver.	13.3	12.1 (90% of Check)		13.9	9.9 (71% of Check)		17.4	10.8 (62% of Check)	

[a] Relative level based on numbers recovered after 45 days: + = increase; - = decrease.

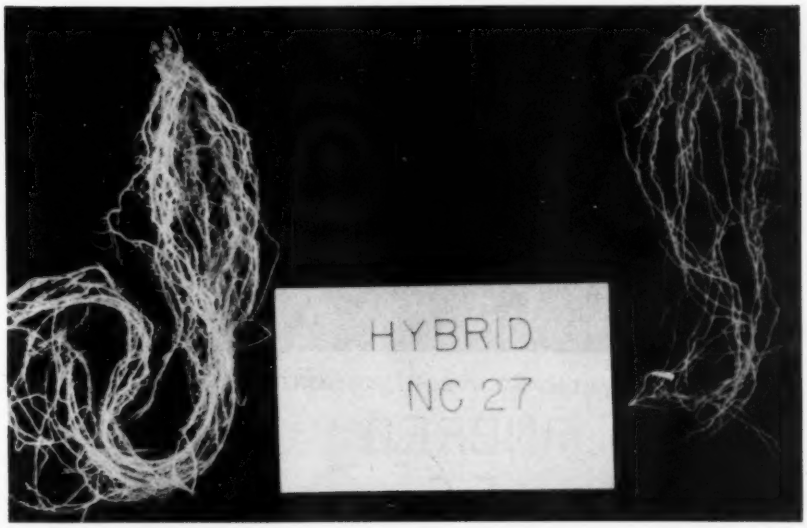

FIGURE 1. Reaction of a susceptible hybrid of corn to the stunt nematode. Left -- check; right - plant grown in infested soil.

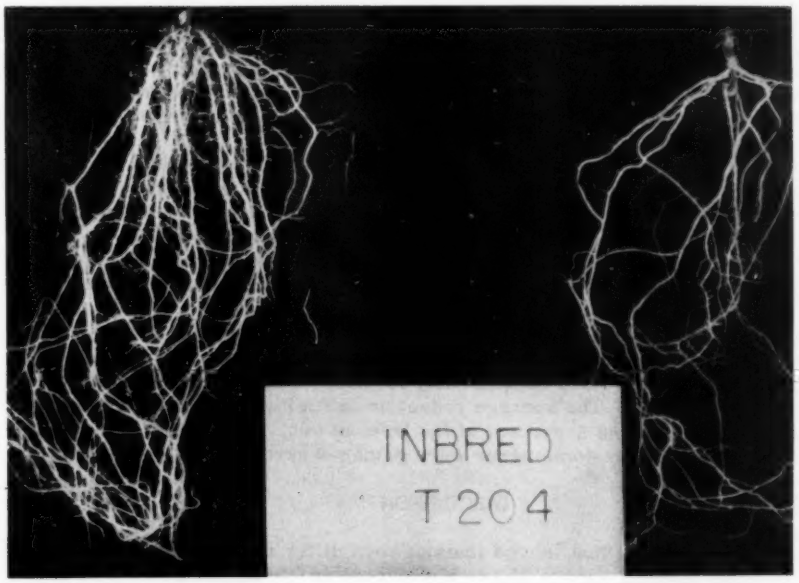

FIGURE 2. Reaction of a susceptible inbred of corn to the stunt nematode. Left -- check; right - plant grown in infested soil.

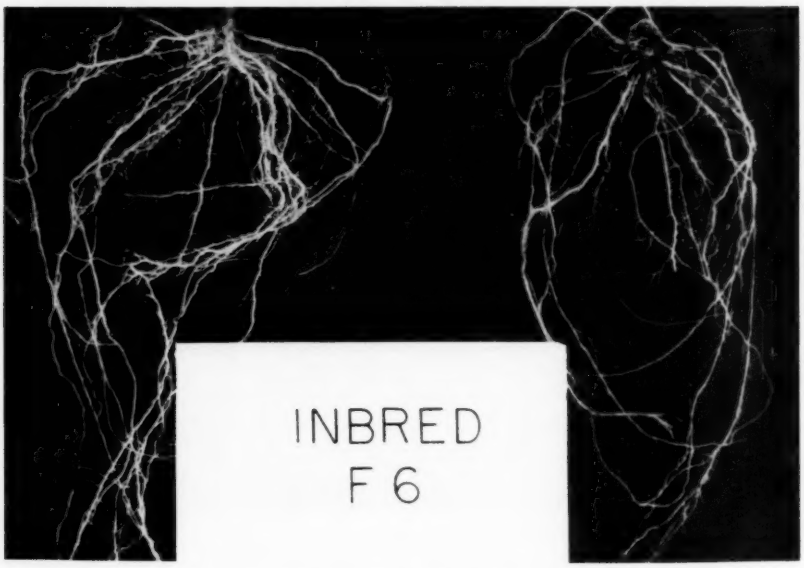

FIGURE 3. Reaction of a resistant inbred of corn to the stunt nematode. Left -- check; right -- plant grown in infested soil.

The 9 resistant inbreds, 5 susceptible inbreds and 3 susceptible single crosses were tested further in soils infested with known numbers of the stunt nematodes. The soils were infested with 50, 200, or 500 nematodes per pot, the number being determined by calculating the number present in a given volume of infested soil and then adding the proper volume of infested soil to each pot. At the conclusion of each test the nematodes from each pot were recovered by the Baermann funnel technique and the populations determined. Root weights also were obtained.

The population of nematodes increased under all inbreds in Group A and all single crosses in Group C (Table 1). However, a reduction in root weight, which was associated with the general appearance of the root systems, was marked only at the higher population levels. The average reduction in root weight for all inbreds in Group A was 54 percent at the level of 500 nematodes, 35 percent at the level of 200 nematodes, and 0 at the level of 50 nematodes per pot. Single crosses (Group C) also suffered greater losses in root weights at the higher population levels.

The number of nematodes decreased under all inbreds in Group B at all population levels (Table 1). There was some reduction in the average weight of roots at all three levels of infestation, although the losses were considerably less than those of inbreds in Group A and the single crosses in Group C. The average reduction in root weight for all inbreds in Group B was 6 percent, 12 percent, and 5 percent at the level of 500, 200, and 50 nematodes, respectively. Individual root systems never were damaged severely.

DISCUSSION

These studies demonstrate that inbred lines of corn differ in their reaction to the tobacco stunt nematode. Some inbreds (Group A) are highly susceptible as evidenced by reduced root systems and by the fact that nematodes reproduce freely when in contact with these inbreds. The populations of the stunt nematode decreased when in association with certain inbreds (Group B) which indicates that they have a certain degree of resistance. Their resistance apparently is not complete because the root systems of all inbreds were damaged to some extent.

High populations of nematodes must be present when corn is in the seedling stage to cause appreciable damage, even to susceptible lines, although reproduction occurs freely. Consequently little or no damage was evident in susceptible inbreds and single crosses when exposed to only 50 nematodes per pot. Corn plants apparently can withstand high populations

of nematodes as the plants become older and develop more extensive root systems.

These data offer a possible indication of the inheritance of resistance in corn to the stunt nematode. Single crosses F6 x F44 and NC7 x T204 are the product of crosses between resistant and susceptible inbreds (Table 1). Since both single crosses are susceptible, as shown by the fact that nematodes reproduce freely on roots of each and the root weights of infected plants are reduced appreciably, it appears that susceptibility is dominant to resistance.

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DEPARTMENT OF PLANT PATHOLOGY, NORTH CAROLINA STATE COLLEGE

WHEAT SNOW MOLD IN EASTERN WASHINGTON 1955 TO 1956[1]

Roderick Sprague[2]

Typhula spp., and associated Fusarium nivale, were severe on fall-seeded winter wheat in Douglas County and to a lesser extent in Okanogan, Lincoln, Chelan, Grant, and Spokane Counties, Washington, during the past winter. A combination of poor dry seeding conditions in the fall, a long winter with continuously deep snow, and some severe freezes in February resulted in almost complete elimination of the fall seedings in Douglas County. Snow fell on unfrozen ground on November 11, 1955, and remained without interruption until mid-April. In fact, at this writing, April 19, snow still lingers in places on Dyer Hill in Douglas County. Wheat began showing decay in December and by February was heavily diseased. By early April it was gone.

Table 1. Winter survival in snow mold plots, April 13, 1956.
Jim Petersen Ranch, Mud Springs, Douglas County, Washington.

TREATMENT	DATE APPLIED	RATE APPLIED PER ACRE	PER CENT STAND			AVERAGE
PCNB (75%) (Mathieson)	October 17	3 lbs. in 40 lbs. Nugreen	Tr.	2	5	2.3
Unsprayed			0	0	0	0.0
Ceresan (M2X) For Snow Mold (Dupont)	October 17	2½ lbs. in 40 lbs. Nugreen	32	30	40	34.0
Nugreen Only (Dupont)	October 17	40 lbs. Per Acre	2	2	1	1.6
Unsprayed			2	2	Tr.	1.3
Captan (Cal. Spray)	October 17	5 lbs. in 40 lbs. Nugreen	8	6	1	5.0
Tag (Cal. Spray)	October 21	3 Pints	12	14	15	13.6
Unsprayed			0	0	0	0.0
Maneb (Rohm & Haas)	October 21	5 lbs. in 40 lbs. Nugreen	0	0	0	0.0
Merbam (Chipman)	October 21	3 lbs. in 40 lbs. Nugreen	Tr.	1.0	Tr.	0.3
Unsprayed			0	0	0	0.0
Ceresan (M2X) For Snow Mold	October 21	2½ lbs. in 40 lbs. Nugreen	30	-	-	30.0
Dinitrobenzene -thiocyanate (Penn Salt)	October 27	5 lbs. in 40 lbs. Nugreen	0	Tr.	2	0.6
PCNB 75%	October 27	3 lbs. in 40 lbs. Nugreen	1	2	0	1.0
Mema (Chipman)	October 27	2½ Pints	45	40	10	31.6
Unsprayed			0	0	0	0.0
Nugreen Only (Dupont)	October 27	40 lbs. Per Acre	0	-	-	0.0
Ceresan (M2X) For Snow Mold	November 8	2½ lbs. in 40 lbs. Nugreen	40	45	55	46.6
Unsprayed			0	0	0	0.0
Captan	November 8	5 lbs. in Nugreen	5	5	5	5.0

[1] Washington Agricultural Experiment Stations Project No. 796.

[2] Plant Pathologist, State College of Washington, Tree Fruit Experiment Station, Wenatchee, Washington.

SUMMARY (All Dates)

TREATMENT	RATE PER ACRE	AVERAGE STAND
Ceresan (M2X)	2½ lbs. in 40 lbs. Nugreen	36.9
Mema	2½ Pints	31.6
Tag	3 Pints	13.6
Captan	5 lbs. in 40 lbs. Nugreen	5.0
PCNB	3 lbs. in 40 lbs. Nugreen	1.6
Dinitrobenzene -thiocyanate	5 lbs. in 40 lbs. Nugreen	0.6
Nugreen Only	40 lbs. Per Acre	0.8
Merbam	3 lbs. in 40 lbs. Nugreen	0.3
Maneb	5 lbs. in 40 lbs. Nugreen	0.0
Unsprayed		0.2

The snow mold plots were situated on the Jim Petersen place near Mud Springs in Douglas County. Last year there was only a trace of snow mold, but this year the locally excellent fall-emerged stand of "Turkey" wheat was destroyed by April 2. The entire region was reseeded to spring wheat in later April and May. Fortunately, moisture conditions were excellent, since the snow melted gradually and water run-off was far less than expected. The snow in January had been nearly 4 feet deep on the plots and in February was about 40 inches deep. In early February we found by digging through the snow that one plot treated with Ceresan M-2X showed about 70 percent survival, one treated with PCNB about 10 percent, and one treated with Captan about 20 percent. By the time final counts were made on April 13, only the Ceresan and Mema-treated plots (Table 1) showed any appreciable survival. As shown in the Table, some plots of Ceresan still had as much as 55 percent of a stand. This, from past experience, should likely result in a sufficiently good stand to warrant retaining. Some Ceresan-treated plots were poorer, especially some treated in October.

Most of our plots were 1/20 acre, replicated. The chemicals (3 to 5 pounds) were applied on the emerged grain with Nugreen-pelleted fertilizer which was used at the rate of 40 pounds per acre. Some of the liquids, such as Tag or Mema, were applied on the emerged wheat in water only in 1/40 A plots, replicated.

Owing to the early heavy snow, only one grower in the area applied Ceresan M-2X on his fields, using 2 1/2 pounds per acre applied by plane in a dust carrier. When County Agent Al Estep and I examined these airplane-treated fields on April 10, there was still 1 foot of wet snow on most of the treated fields. The white wheat had winter-killed too badly to determine very much. Snow mold was not prevalent. The Turkey-type wheat had also winter-killed but still showed about 30 percent survival. Winter injury therefore confused the issue in these plane-dusted fields.

This year the cost of basic mercury has been reflected in the increased cost of mercurial fungicides. Under current retail prices, Ceresan M-2X (Dupont) and Mema (Chipman) cost over $5.00 per acre. Under present economic conditions this is too expensive for most growers in the Douglas County area. However, the treatment is being used commercially in some areas in Okanogan County, with expanded acreage planned for treating next year. In the community of Havillah, which lies at an elevation of 3500 feet, winter wheat consistently is destroyed by mold unless treated. Here we found both Ceresan M-2X and Liquid 365 (Dupont) used. I saw one field treated with 3 pints of Liquid 365 that had 100 percent stand, whereas untreated fields showed less than 5 percent stand in late April. The growers had tried Ceresan M-2X at 1 1/4

pounds per acre (half recommended strength) and Liquid 365 at 1 1/2 pints. In 1956, stand was about 45 to 60 percent, and the growers planned full strength treatment next fall. They prefer the Liquid 365 applied with a weed sprayer with enough water to give complete coverage.

In the Havillah area spring wheat is too late to be practical. Without chemical treatment, winter wheat cannot be grown during most years. In fact, until the treatments were tried, winter wheat had been abandoned in the area. Since a cash grain crop bolsters the local economy, the growers are willing to pay the $5.00 plus costs per acre. Their approval of a liquid material calls for increased study of all available liquid materials in next year's trials.

The aid of Country Agent Al Estep of Douglas County during the winter is appreciated. Officials of the Dupont Company who furnished a grant-in-aid during earlier seasons are thanked for their cooperation.

STATE COLLEGE OF WASHINGTON, TREE FRUIT EXPERIMENT STATION WENATCHEE, WASHINGTON.

THE REACTION OF SPECIES OF POA AND OTHER GRASSES TO PUCCINIA STRIIFORMIS[1]

Michael P. Britton and George B. Cummins[2]

In conjunction with a study of stem rust of bluegrass, a sample of a bright yellow rust of Merion bluegrass was received from Dr. Malcolm A. Nobs, Carnegie Institution of Washington,

Table 1. Reaction of bluegrasses to a culture of Puccinia striiformis isolated from Poa pratensis.

Species	Line[a]	Source	Reaction
Poa ampla Merr.	CL-4173-1	Condon, Oregon	S
" "	CL-4178-1	Kahlotus, Washington	S
" "	CL-4183-1	Albion, Washington	S
" "	CL-4196-12	Heise Hot Springs, Idaho	R
" arachnifera Torr.	CL-4248-13	Stillwater, Oklahoma	S
" canbyi (Scribn.) Piper	P-851	SCS Nursery, Pullman, Washington	S
" compressa L.	CL-4260-1	Chorsum, Turkey	S
" "	P-410	SCS Nursery, Pullman, Washington	S
" "	PU-5298	PI 182794	S
" nemoralis L.	PU-5299	PI 182858	S
" pratensis L.	PR-6131	Washington State College	R
" "	PR-6132	" " "	R
" "	PR-6133	" " "	R
" "	PR-6134	" " "	R
" "	PR-6135	Minnesota 95	S
" "	PR-6136	clonal selection, Birch Point, Canada	S
" "	PR-6137	Kansas State College	S
" "	PR-6138	" " "	S
" "	PR-6139	" " "	S
" "	PR-6140	" " "	S
" "	PR-6141	" " "	S
" "	PR-6142	PI 196331	I
" "	PR-6143	PI 198075	S

[1] This investigation was conducted as part of a project supported by the Turf Research Foundation.

[2] Instructor of Plant Pathology; and Professor of Plant Pathology, Purdue University and Agent, Plant Disease Epidemics and Identification Section, United States Department of Agriculture.

Stanford, California. The fungus, because of the morphology of the urediospores and teliospores, as well as the seriate arrangement of the uredia in long, narrow, chlorotic streaks, was identified as Puccinia striiformis West. (P. glumarum Eriks. et E. Henn.). Since stripe rust has not been reported previously on bluegrasses, inoculation experiments were carried out. A series of grasses was inoculated in the greenhouse using inoculum from freshly sporulating infections on Merion bluegrass. No infection resulted on Arrhenatherum elatius (L.)

Table 1. (Continued).

Species	Line[a]	Source	Reaction
Poa pratensis L.	PR-6145	PI 204485	I
" "	PR-6146	PI 204486	S
" "	PR-6148	PI 204487	R
	PR-6149	commercial, Iowa	S
" "	PR-6150	" , Minnesota	S
" "	PR-6151	" , North Dakota	S
" "	PR-6152	" , Kentucky	S
" "	PR-6153	" , Missouri	S
" "	PR-6154	Merion	S
" "	PR-6155	Delta	S
" "	PR-6156	PI 119684	S
" "	PR-6157	PI 198075	S
" "	PR-6159	commercial, England	S
" "	PR-6160	clonal selection, Illinois	S
" "	CL-4249-1	Athabaska, Canada	I
" "	CL-4553-4	Mather, California	S
" "	CL-4466-1	Newport, Oregon	S
" "	P-4358	SCS Nursery, Pullman, Washington	S
" "	PU-5288	S-2 Marysville, Ohio	S
" "	PU-5289	S-4 Marysville, Ohio	S
" "	PU-5290	K-34 Pennsylvania State University	S
" "	PU-5291	Merion (Breeders seed)	S
" "	PU-4509	Selection from Chicago, Ill.	S
" scabrella (Thurb.) Benth.	CL-4212-1	Las Posas, California	S
" "	CL-4214-1	Watsonville, California	S

[a] P = accessions of the Pullman Nursery Unit of the U. S. D. A. Soil Conservation Service.
CL = line numbers of the Carnegie Institution of Washington, Stanford, California.
PU = accessions of Purdue University, Agronomy Department.
PR = 1956 row numbers Purdue University, Agronomy Department.

Presl, Bromus marginatus Nees, B. tomentellus Boiss., Festuca ovina L., F. ovina var. duriuscula (L.) Koch, Koeleria cristata (L.) Pers., Poa annua L., P. caespitosa Forst., P. iridifolia Hauman, and one selection of Alopecurus pratensis L. Alopecurus arundinaceus Poir. and one selection of A. pratensis were susceptible. The reactions of other species and selections of Poa are recorded in Table 1.

The reaction types in Table 1 were arbitrarily determined as follows: I = immune, no sori developed; R = resistant, considerable necrosis, sparse and small uredia; S = susceptible, no necrosis, abundant and often coalescing uredia.

These data are presented because of the current interest in disease resistance improvement of bluegrasses and because we do not intend to continue the study. It is improbable that stripe rust will be important in the Midwest, but it may be found to be important in the Western States where favorable temperatures prevail.

PURDUE UNIVERSITY, LAFAYETTE, INDIANA, AND UNITED STATES AGRICULTURAL RESEARCH SERVICE

A BACTERIAL-INDUCED CHLOROSIS IN THE LEE SOYBEAN

Lewis W. Erdman, Herbert W. Johnson, and Francis Clark

The Lee soybean, bred for mid-southern adaptation and for other selected qualities, was released to the public in 1954. It was anticipated that the variety would find ready acceptance and that it would largely replace Ogden, which shatters readily, has a green seed coat, and is highly susceptible to purple seed stain.

During the initial and in subsequent crop years, growers of Lee variously noted and objected to an upper-leaf chlorosis appearing during the pre-bloom stage. The condition was spotty within fields. The number of plants with yellowish-white top leaves ranged from negligible percentages to as high as 10 percent at any one time. Many of the afflicted plants recovered spontaneously within a week or so. Meanwhile, of course, others might develop chlorosis. The malady appeared in all areas where Lee was grown; it appeared somewhat more prevalent on sandy than on heavier-textured soils. The extent of yield reduction if any has not been defined. Because the majority of the plants are not affected, while many of the chlorotic plants do recover, we believe that any effect on yield is slight. Additional information is needed before any more positive statement on crop damage can be made.

In the course of strain testing of rhizobia in field plots of Lee soybeans at Brawley, California, in 1954 and 1955, in work designed primarily for selecting nitrogen-fixing bacteria for use in the Southwest, a similar irregular chlorosis was noted. The chlorosis appeared to be associated with certain rhizobial inoculants. In order to investigate more fully the etiology involved, Lee soybeans were grown in greenhouse cultures at Beltsville, Maryland, during 1955 and 1956. The plants were grown in a sand-nutrient solution substrate. The nutrient solution, minus nitrogen, was a modified Crone's solution that heretofore has been found suitable for culturing legumes in the course of greenhouse testing of rhizobia. Seeds were surface-sterilized and then inoculated with various pure cultures of rhizobia. Some of these cultures were selected because of their being suspect in the Brawley plots; others added in later tests were isolated from root nodules present on chlorotic plants.

Altogether, four separate experiments, with all inoculant treatments at least in triplicate, were conducted with Lee soybeans in sand substrates. Upper leaf chlorosis, varying from light green to nearly white leaves, regularly appeared in all plants given certain inoculants and in no plants given other inoculants. In many cases vegetative yields of the chlorotic plants at the time of blooming were as good as or even better than those of plants inoculated with highly effective rhizobia which did not produce any chlorosis. Good nodulation was observed on roots of the chlorotic plants. The nodules were definitely not of the parasitic type. They were large and numerous and clustered around the tap root. With severe chlorosis there was stunting of plant growth and reduction in yield.

Work is now in progress to study the susceptibility of other soybean varieties to this bacterial-induced chlorosis and to define more fully the conditions affecting its incidence and severity in the Lee soybean.

AGRICULTURAL RESEARCH SERVICE, UNITED STATES DEPARTMENT OF AGRICULTURE, BELTSVILLE, MARYLAND

COORDINATION IN BREEDING TOMATOES FOR RESISTANCE TO CLADOSPORIUM LEAF MOLD[1]

Emil F. Guba

Summary

A long-time tomato breeding program has ultimately produced red forcing tomatoes highly resistant to and immune from Cladosporium leaf mold (Cladosporium fulvum). The resistant and immune ancestors in the pedigree of the Waltham tomatoes are primitive species of Lycopersicon native to the Andean region of western South America. Our finished tomatoes run 3 to 5 to the pound; their South American ancestors 450 to the pound.

Resistance to and immunity from Cladosporium leaf mold are dominant genetic characters. Advantage is shown by crops from F_1 hybrid seed combining disease resistance and high productivity, e.g., Improved Bay State (resistant) or Waltham Mold Proof Forcing (immune) x either Waltham Forcing or Michigan State Forcing (both completely susceptible).

The success of the effort is due to the Lycopersicon accessions from the Section of Plant Introduction, United States Department of Agriculture, and the contributions of Federal and State agencies and plant pathologists, all of whom are duly recognized in the text.

Our work has brought advantage and security to the tomato forcing industry in New England.

The breeding of red forcing tomatoes for resistance to Cladosporium leaf mold (Cladosporium fulvum) in Massachusetts began in 1934, following a report on disease resistance in tomatoes by Sengbusch and Loschakowa-Hasenbusch (9). The effort has been mixed with disappointment, frustration, and ultimate success. The cooperation of State and Federal agencies and professional colleagues throughout the period of study is noteworthy.

Bay State Forcing, the first of the Massachusetts tomato varieties to result from these studies, derived its immunity from the original pathogenic races of tomato leaf mold from a seed store packet of Lycopersicon pimpinellifolium L. The tomato was accepted in commercial culture; then suddenly showed complete susceptibility to the pathogen (5). We were faced with the problem of new virulent races of Cladosporium.

Sample quantities of seed of many primitive types of tomatoes were obtained from the Regional Vegetable Breeding Laboratory, United States Department of Agriculture, Charlestown, South Carolina, and from Dr. L. J. Alexander, Ohio Agricultural Experiment Station. The plants were appraised for their reaction to a composite collection of Cladosporium leaf mold in New England. Several types were immune, among them two types of Lycopersicon pimpinellifolium with smooth red fruits, namely U. S. D. A. Pl. Intr. no. 112215 with fruits 1/2 inch in diameter, from Ecuador (Fig. 3A), and no. 129882 with fruits nearly 1 inch in diameter, from Peru. After a period of breeding for commercial type, the lines with the no. 129882 ancestor were lost to additional races of the pathogen. No. 112215 remained immune, and from it came several lines of tomatoes with distinct pedigree, each given a distinct designation (6). The best line, designated Improved Bay State, was released in 1946: (Bay State x Pl. Intr. 112215) x Waltham Forcing (Fig. 4B).

In 1950 Bailey (2) recognized 7 known pathogenic races of Cladosporium and reported Pl. Intr. no. 112215 immune from all of them. Similar circumstances have been reported by Day (4). Kerr (7) placed Pl. Intr. 112215 in the non-sporulating resistant classes 1 and 2, with 1 implying no disease, 2 high resistance.

The Waltham tomatoes and varieties from other sources were tested in 1945 and 1946 in Peru, South America, where U. S. D. A. Pl. Intr. no. 112215 is native (3). In these tests Bay State was completely susceptible. Improved Bay State was outstanding in resistance and performed the best. This variety averages 3 to 5 tomatoes to the pound. It has a wide range of commercial culture today. In the course of study Improved Bay State lost its immunity from infection and instead now shows high resistance typical of the non-sporulating resistance class 2 (7).

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[2]Contribution No. 1049 of the University of Massachusetts, College of Agriculture, Massachusetts Agricultural Experiment Station, Amherst, Massachusetts.

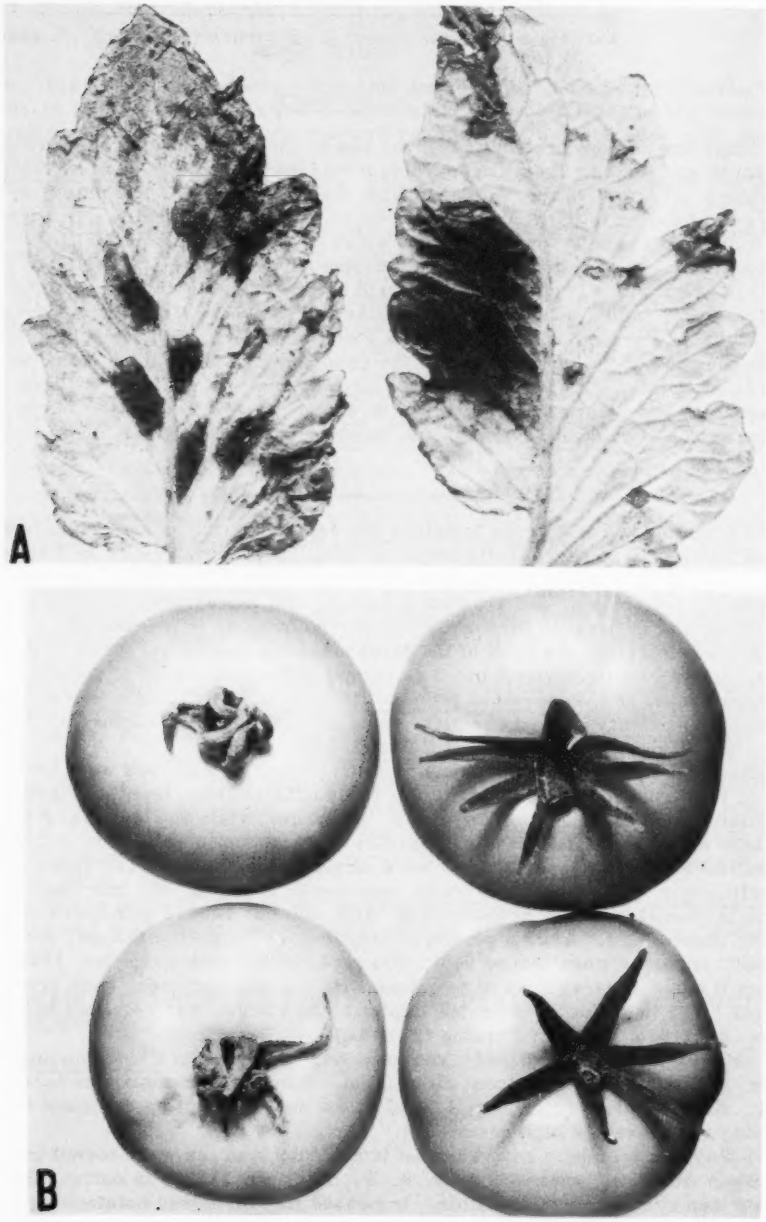

FIGURE 1. A -- Lower side of tomato leaflets showing diseased areas infected with Cladosporium leaf mold (from Florida Agr. Exp. Sta. Bull. 185, Fig. 84, 1926). B -- Calyx on fruit at left destroyed by pathogen; right, healthy and green.

FIGURE 2. Segregation for complete susceptibility to and immunity from Cladosporium leaf mold.

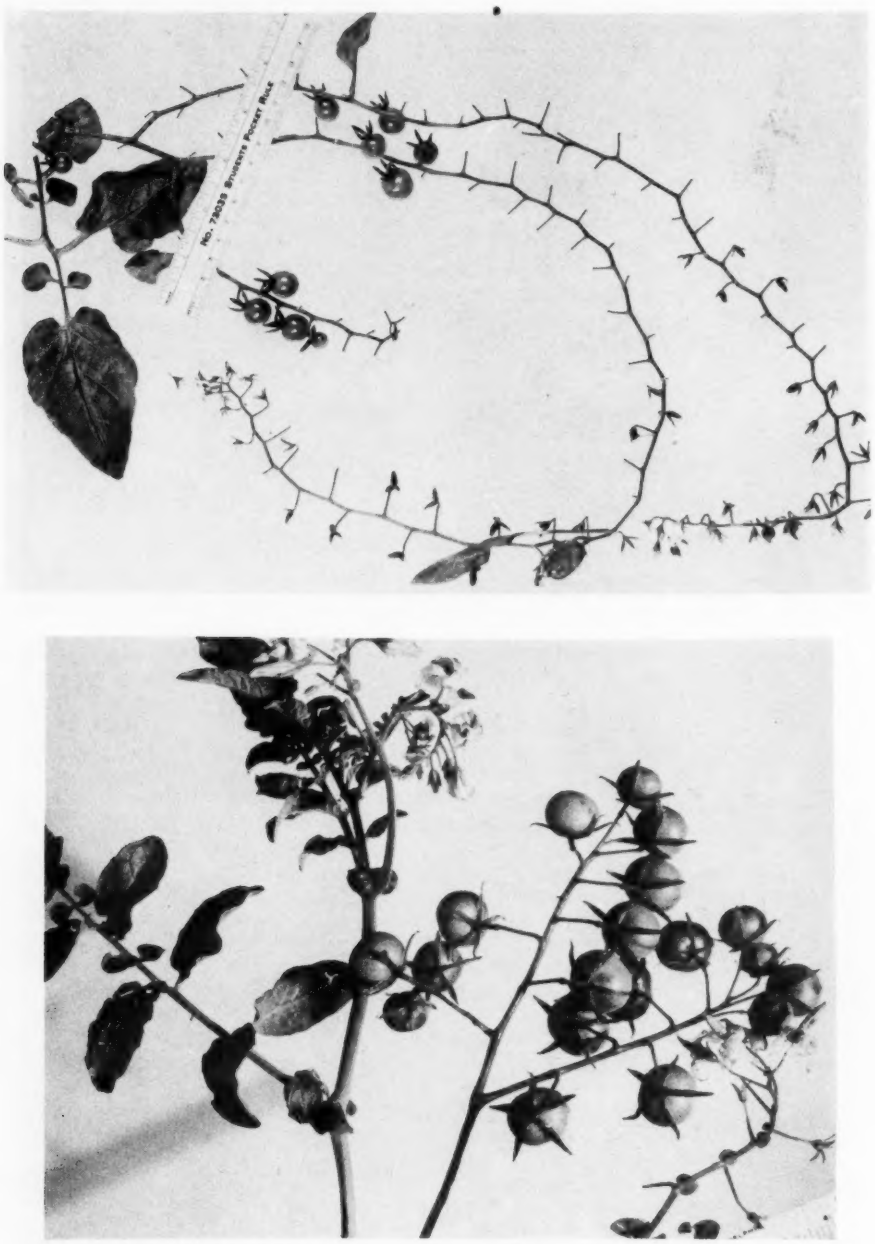

FIGURE 3. A --Red Currant tomato, Lycopersicon pimpinellifolium L., U.S.D.A. Pl. Intro. No. 112215 in the pedigree of Improved Bay State. B -- Peruvianum tomato, Lycopersicon peruvianum L., in the pedigree of Porte and Walker 44B292 and Waltham Mold Proof Forcing.

FIGURE 4. Foliage reactions to infection by Cladosporium fulvum. A -- completely susceptible; Waltham Forcing; B -- highly resistant, Improved Bay State; C -- immune, Waltham Mold Proof Forcing. Similar reactions to infection appear among F_1 hybrids of these tomatoes x completely susceptible varieties.

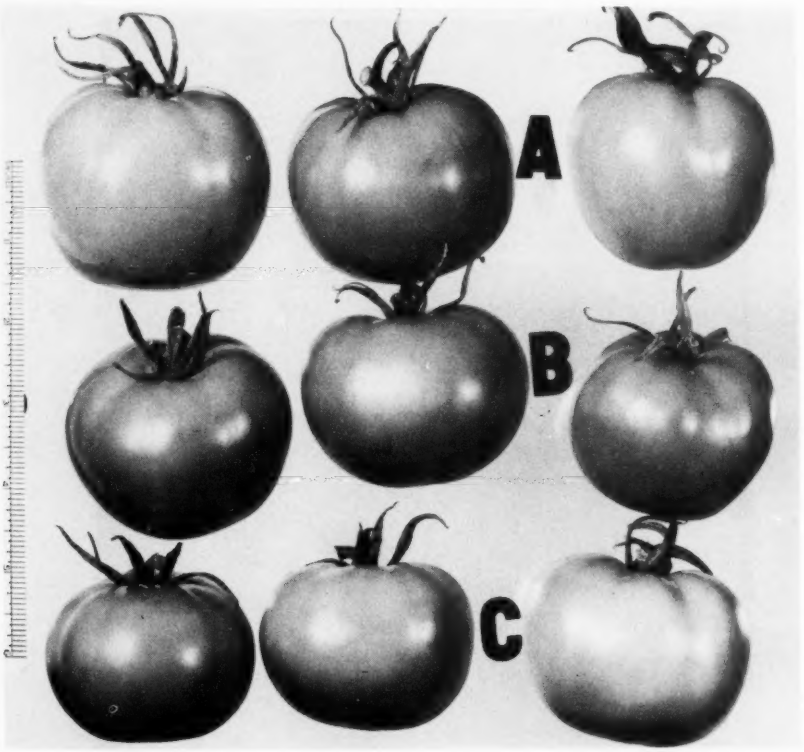

FIGURE 5. A -- Fruit of Ontario V-500 Series x Waltham Mold Proof Forcing featuring deep globose fruit of the Vineland parent. B and C -- Waltham Mold Proof Forcing featuring globose oblate fruit.

Several accessions of Lycopersicon peruvianum (Fig. 3B), another Andean tomato, were found to be totally immune from composite races of the fungus in New England (5), but efforts to cross this species with L. esculentum always failed. Porte and Walker (8) succeeded in making this interesting cross. An F_2 line was crossed with Pan America and designated U. S. D. A. 44B292. These tomatoes had no commercial value. Stable lines were developed at Waltham for immunity and type, and from this material Waltham Mold Proof Forcing 2-22, immune from all races of the pathogen was developed: (U. S. D. A. 44B292 x Improved Bay State) x Improved Bay State. Waltham Mold Proof Forcing 2-22 was crossed with Waltham Forcing, stabilized, and designated Waltham Mold Proof Forcing 54. The difference between the two is trivial. The Waltham Mold Proof Forcing tomatoes run 3 to 4 to the pound. Commercial culture has given preference to no. 2-22. The variety is extensively grown under glass in New England (Fig. 4C).

Immunity appears in numerous breeding lines. Series Vineland-500 developed by Dr. E. A. Kerr, Ontario Horticultural Experiment Station, is derived from L. hirsutum, L. pimpinellifolium (Pl. Intr. no. 112215), and other sources. These tomatoes have a pleasing globose form but for other reasons, according to our grower tests, are not satisfactory. Lines of V-500 were crossed with Waltham Mold Proof Forcing (Fig. 5) and a line designated Waltham Mold Proof Forcing 56 is under commercial trial.

Resistance and immunity are dominant factors and feature the F_1 progeny of hybrids of the Waltham tomatoes x completely susceptible varieties. Therefore F_1 hybrids, e.g., Improved Bay State or Waltham Mold Proof Forcing x Michigan State Forcing, combining disease resistance, hybrid vigor, and high productivity, are being promoted in the New England area.

Also, by selection and further hybridization it should be possible to develop field or other forcing types acceptable to other geographic areas where leaf mold is a problem, or to add resistance to other diseases.

The annual production of forcing tomatoes around the Boston area is about 1200 tons, representing a gross income of nearly $700,000.00. Competition with shipped-in tomatoes from distant producing areas is considerable. Our study has contributed a measure of security to our tomato forcing industry. With the change to these new tomatoes the devastation from Cladosporium leaf mold common in the past no longer occurs.

Our breeding work was always done in the presence of an epidemic of tomato leaf mold. Promising lines were released as soon as the fruit and yield appeared satisfactory, even though the lines were not pure for resistance or immunity or commercial type. Thus we have been privileged to have the guidance of our tomato forcing industry throughout the progress of our long effort toward our ultimate objective.

The Cladosporium resistant and immune accessions (used in our studies) do not possess horticultural features peculiar to forcing tomatoes as alleged by Andrus (1). These primitive species are odd; the fruits average 450 to the pound, and like field tomatoes, vine strongly and set fruit poorly in greenhouse culture.

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PRELIMINARY TESTS INDICATE THAT THE SMALL OAK BARKBEETLE MAY BE A VECTOR OF THE OAK WILT FUNGUS

William D. Buchanan[1]

In Missouri, the small oak barkbeetle Pseudopityophthorus minutissimus (Zimm.), breeds in most scarlet and black oak trees that develop the oak wilt disease. In many instances this beetle begins its attack about the time the first foliar symptoms of the disease are observed, and it may continue to attack until the cambium is more or less dry and brown. Some of the beetles that emerge from diseased trees may breed to some extent in live branches that are being "shaded out" on healthy trees. They may also do a limited amount of feeding in twig crotches throughout the crowns of healthy trees. These and other habits indicate that, under favorable conditions, this insect might be involved in the spread of the oak-wilt fungus Endoconidiophora fagacearum Bretz from diseased to healthy trees. On this assumption, the following study was made.

Materials and Methods

Several thousand P. minutissimus were reared from sections of oak-wilt-killed trees in the spring and summer of 1954. These insects were macerated in water, and the resulting mixture was injected via ax or knife cuts into the xylem of 74 healthy scarlet and black oak trees. The trees ranged in size from about 1/2 to 8 inches in diameter. Injections were made between May 20 and September 11, 1954 in oak trees growing in a natural stand on the Sinkin Experimental Forest near Salem, Missouri.

The following checks were used in this study: 1) Eighteen trees were inoculated with several thousand beetles macerated in a suspension of oak-wilt fungus spores; 2) fifteen trees were inoculated with a mixture of water and several thousand macerated beetles that had emerged from nondiseased trees; 3) several hundred other trees in the study plot were not inoculated.

Results

No recognizable oak-wilt symptoms developed in any of the trees in 1954. In 1955 foliar symptoms developed in 6 of the 74 trees that were inoculated with macerated beetles that emerged from oak-wilt-killed trees. Symptoms also developed in 6 of the 18 trees that were inoculated with macerated beetles in a suspension of oak-wilt fungus spores. No symptoms developed in any of the 15 trees that were inoculated with a mixture of water and macerated beetles that emerged from nondiseased trees or in any of the trees that were not inoculated. The oak-wilt fungus was isolated from all trees that developed foliar symptoms of the disease[2]

Conclusions

Some of the small oak barkbeetles that emerge from oak-wilt trees carry the oak-wilt pathogen in or on their bodies. Similar conclusions were reached by Stambaugh et al.[3] who isolated the oak-wilt fungus from beetles that emerged from diseased wood in Pennsylvania. The Missouri tests demonstrated that the beetles carried sufficient amounts of the pathogen to cause healthy trees to get oak wilt after they were inoculated with a mixture of the macerated beetles in water. However, before the beetles can be considered as agents in the spread of oak wilt, it will have to be shown that they can complete the final step -- namely, introduce the pathogen into infection courts in healthy trees.

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[1] Forest Entomologist, Central States Forest Experiment Station, Forest Service, United States Department of Agriculture, Columbia, Missouri. Contribution from the Central States Forest Experiment Station, Forest Service, United States Department of Agriculture, and Missouri Agricultural Experiment Station, Journal Series No. 1627. Approved by Director.

[2] Cultures and determinations of the fungus were made by Dr. T. W. Bretz, Forestry Department, University of Missouri.

[3] Stambaugh, W. J., W. J. Fergus, F. C. Craighead, and H. E. Thompson 1955. Viable spores of Endoconidiophora fagacearum from bark and wood boring beetles. Plant Dis. Reptr. 39: 867-871.

ELSINOË AND SPHACELOMA IN FLORIDA. II

Albert S. Muller[1]

Recent new discoveries of Elsinoë and Sphaceloma in Florida are as follows:

ELSINOË LEDI (Peck) Zeller (9)

On Leucothoe populifolia (Lam.) Dipp., leaves, Middleburg, January 15, 1956. A. S. M. (F 44962)[2]

New host genus and species, also new distribution. Previously recorded on Ledum spp., northern and western United States (Maine, New York, Pennsylvania, Michigan, Minnesota, Washington, Oregon, California) (4, 9), also Canada (3), and on Gaultheria shallon Pursh., Washington (6).

ELSINOE LEUCOSPILA Bitanc. & Jenkins (2)

On Ternstroemia gymnanthera Sprague, leaves, Jacksonville Beach, March 10, 1956. A. S. M. (F 45054).

New host genus and species. Formerly recorded from Florida on Camellia spp. (7, 8), as well as provisionally on C. sinensis (L.) Kuntze (2).

SPHACELOMA PSIDII Bitanc. & Jenkins (1)

On Feijoa sellowiana Berg., leaves, Gainesville, April 7, 1956. A. S. M. (F 45053).

Previous records on this host are from New Zealand, also from southern Brazil (5). S. psidii was described on Psidium guajava L. from São Paulo, Brazil.

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[2]Accession number, Herbarium, Florida Agricultural Experiment Station, Gainesville, Florida.

PLANT DISEASES OBSERVED IN EL SALVADOR DURING SUMMER, 1955[1]

Leopoldo Abrego, Bernardo Patiño, and George F. Weber[2]

During the summer of 1955, observations made and experimental work conducted in El Salvador were conducive to the compilation of the following list of parasitic fungi causing diseases of cultivated plants, with special emphasis on those of particular economic importance. This report in no way constitutes a plant disease survey of the area, as has been previously published (1, 2). Immediately following will be some pertinent observations, after which there will be an alphabetized list according to the common Spanish name of the host plants followed by their Latin binomial, the name of the causal organism, and the name of the disease in Spanish.

ALTHAEA, Hibiscus syriacus, occasionally found in scattered places as an ornamental shrub, was usually affected with rust (Kuehneola malvicola (Speg.) Arth.) which caused various stages of defoliation, frequently complete.

ANONA, Annona reticulata: About 50 percent of the fruit of this tree was decaying from the stem-end rot caused by the fungus Diplodia natalensis Pole-Evans. In most cases, fruits were shed from the trees before maturity.

AVOCADO, Persea americana, was found affected with most of the common diseases associated with this plant, scab (Sphaceloma perseae Jenkins) being most common. The disease was very prevalent on the foliage and mature fruit although it was not causing defoliation. Probably the second most frequently found disease was the leaf-spot, Cercospora purpurea Cke.

BANANA, Musa paradisiaca sapientum, which grows luxuriantly, both in cultivation and as volunteer plants, was frequently observed affected with the leaf-spot fungus Cercospora musae Zimm. The disease in no instance appeared to be of such proportions as to be considered detrimental to the development of the host plant or the production of fruit.

CANNA, Canna indica: Wherever this plant was grown, both in cultivation and escaped, it usually was quite heavily infected with the uredinial stage of Puccinia cannae (Wint.) P. Henn. In certain instances the rust was severe enough to prevent normal development of the plant.

CHRYSANTHEMUM, Chrysanthemum morifolium, grown extensively during this period of the year, was almost without exception affected by Septoria chrysanthemi Allesch. Usually the disease was not severe.

COFFEE, Coffea arabica L., is the most commonly cultivated plant in this country. It is affected with several important diseases of which damping-off in the seedbed, caused by Rhizoctonia solani Kuehn, is common. The horticultural practices followed in the development of seedlings, particularly the dense shade and the natural moisture of the rainy season, distinctly favor the development of this fungus, and in many instances the loss of seedlings is excessive.

Leaf-spot, Cercospora coffeicola Berk. & Curt., in some seedbeds was as destructive as the damping-off disease. It was observed on the cotyledons and continued to develop on the foliage of the plants. The disease was so severe in several instances that seedbeds containing 30,000 to 50,000 plants were abandoned. The disease continued to develop upon the young seedlings, causing them to become defoliated when 6 to 10 inches tall. Stem lesions, causing

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[1]A contribution from the Centro Nacional de Agronomia, Santa Tecla, El Salvador, a technical agricultural organization operated jointly by the Governments of El Salvador and the United States. United States participation in this work was carried out as part of the Technical Cooperation Program in El Salvador administered by the International Cooperation Administration.

[2]Respectively, Subchief and Assistant Subchief, Department of Plant Pathology, Centro Nacional de Agronomia, Santa Tecla, and Research Adviser, Plant Pathology, United States Operations Mission to El Salvador.

dieback, were also frequently found on new growth. This disease was also observed on coffee plants in all stages of development and became less severe in proportion to the size and age of the plant. The immature berries were occasionally affected with this fungus. Some control experiments were conducted in the seedbeds by the application of fungicides at specified intervals. These trials indicated that Cuprocide, Manzate, and Copper A controlled the disease sufficiently to insure transplanting stock in contrast to the checks which were killed by the parasite.

Thread-blight, Pellicularia koleroga Cke., was observed in several intensive coffee-growing areas. It was exceedingly destructive in these limited plantings. Frequent and careful observations indicated that the fungus survived the dry season on the aerial parts of the host without the development of sclerotia or sclerotia-like structures that could in any way be distinguished from the well-known mycelial threads commonly observed. The fungus grew luxuriantly under the prevailing summer conditions, followed the branches and new growth from the old wood, invaded the leaf blade tissue from the base of the petiole, eventually killing the leaf which usually turned brown and remained attached at its base by the thread of the fungus. Threads were mostly found on the lower surface of the twigs, foliage, and berries, the latter becoming brown from the invasion of the mycelium in contrast to the healthy green surface of the fruit. Application of fungicides for control of thread blight was inaugurated. The effectiveness of these must, of necessity, depend on the efficiency and frequency of applications correlated with the terrain. Also, satisfactory results will possibly be dependent upon the application of fungicides correlated with spot pruning.

Leaf-spot, Omphalia flavida (Cke.) Maubl. & Rangel, apparently survives from season to season on the older leaves of the host plants that were not sufficiently diseased during the previous season to cause shedding. There may be some possibility also of the fungus surviving on detached leaves on the soil surface. During the rainy season the fungus developed rapidly, and the disease was exceedingly destructive, causing defoliation of the host plant. The disease was usually found to make its appearance on the leaves closer to the soil and rapidly involved areas higher up the plant until, in many instances, the trees were entirely defoliated. The leaf-spots, usually limited to a centimeter in diameter, appeared several to many on a single leaf and caused rapid defoliation. Apparently some product of the fungus causes abscission, whereas similar spots in size and number caused by other fungi did not result in leaf drop. The fungus was found on numerous wild plants growing in the coffee plantings, and, consequently, adequate control required clean cultivation and the application of fungicides to both the host and the soil surface which in most instances was almost impossible. Some preliminary trials with various fungicides offered promising results.

Anthracnose, Colletotrichum coffeanum Noack, was generally distributed in coffee plantations during the middle and latter part of the rainy season. Apparently it spread most rapidly on young immature foliage and young shoots that may have over-extended themselves due to favorable growing conditions. The disease assumed important proportions only rarely even though its distribution was general. It is apparently closely associated with fertility and environmental conditions.

Leaf-spot, Phyllosticta coffeicola Speg., was frequently indistinguishable from the preceding disease in appearance and distribution, and closer examination was necessary to determine the difference. It did not occur frequently although its distribution was more or less general. There was no economic importance associated with it in any particular locality.

Black root-rot, Rosellinia bunodes (Berk. & Br.) Sacc., about which there is no unanimity of opinion among plant pathologists as to its cause, continues to be an important disease of coffee. The disease appeared in definite areas not particularly correlated with elevation or soil types. Diseased plants were distributed in definite group patterns rather than at single and widely scattered locations.

CORN, Zea mays, which is grown extensively, showed a low percentage of rust, Puccinia sorghi Schw., and a trace of smut, Ustilago maydis (DC.) Cda.

Leaf-blight, Helminthosporium turcicum Pass., was prevalent wherever the host plant was grown. Some plantings were much more severely affected than others, and in these instances considerable reduction in yield might be expected. Many different selections of corn growing in close proximity showed considerable variation in susceptibility to this disease. There happened to be no particular correlation between the plants that were growing in marginal areas in contrast to those in well-fertilized and cultivated fields.

COTTON, Gossypium hirsutum, is one of the principal crops in the coastal region of the country, and the principal trouble encountered during the early development of the plants was damping-off, Rhizoctonia solani Kuehn, causing in many instances a 10 to 25 percent reduction in stand. An excessive amount of seed was used to compensate for this loss. Associated with the seedling stage, there also occurred a leaf-spotting disease, Rhizoctonia sp., characterized by defoliation of cotyledons and true leaves in severe cases and by leaf-spotting at other times. The two fungi were distinguishable in culture.

CRAPE-MYRTLE, Lagerstroemia indica, was severely attacked by the powdery mildew fungus, Erysiphe lagerstroemiae E. West, wherever there happened to be a lack of sunshine and air drainage. In such instances, the leaves were malformed and the flower buds failed to develop, remaining blighted and unopened. Only the Oidium stage of the fungus was observed.

Thread-blight, Rhizoctonia ramicola Weber & Roberts, frequently attacked this plant. It caused killing of the foliage, particularly on the new growth beginning from the old wood and extending upward. Frequent and close observation of diseased plants failed to reveal sclerotia although there was an aggregation of aerial hyphae at the base of the petiole.

CUCUMBER, Cucumis sativus, wherever grown, was observed to be affected with downy mildew, Pseudoperonospora cubensis (Berk. & Curt.) Rost. Plants suffered considerably from loss of foliage. Control measures, in most instances, failed to check this disease and reduced production resulted.

Leaf-spot, Phyllosticta cucurbitacearum Sacc., was observed on several occasions on plantings widely separated from each other. In each instance the disease was of greater importance than any other disease affecting the host plant. The fungus was observed sporulating on the host in each case.

DAHLIA, Dahlia pinnata, commonly grown, was usually found affected with leaf smut, Entyloma dahliae Syd. Extensive and often complete defoliation, with a definite stunting of the host, occurred. The disease usually developed near the ground on the older leaves and progressed up the plant on the younger foliage.

EGGPLANT, Solanum melongena, wherever grown, was considerably affected by Diaporthe vexans (Sacc. & Syd.) Gratz, resulting in heavily spotted older leaves, the typical tip-over symptom on the main stem, and the characteristic fruit rot which is most diagnostic of the disease.

Stem-rot, Sclerotinia sclerotiorum (Lib.) D By., was not found frequently. On occasion, the fungus was observed on fruits as well as on the stem at the soil line. Several plants were also observed showing symptoms suggesting virus diseases.

FIG, Ficus carica, as observed, was always infected with the rust, Physopella fici (Cast.) Arth. In most instance, the plants had been defoliated and the new growth was appearing.

GLADIOLUS, Gladiolus hortulanus, was grown by several commercial tradesmen as well as local flower gardeners. In most instances leaf and flower spotting, caused by Botrytis gladiolorum Timm., was observed. This usually limited the keeping quality of the cut flowers.

Red-spot, caused by Stemphylium sp., was prevalent in most plantings. It caused considerable spotting and frequently stunting of the foliage at blossoming time.

Brown spot, attributed to Curvularia lunata (Wakk.) Boed., was observed only on certain varieties in commercial plantings, some of which were so seriously damaged that there would probably be few salable flower stalks cut from the plants.

HIBISCUS, Hibiscus rosa-sinensis, widely cultivated as an ornamental shrub, was frequently found partially defoliated by the silky thread-blight fungus, Rhizoctonia ramicola Weber & Roberts. The threads of the fungus grew up the new growth, invading the leaves and killing the tissue from the petiole up the blade, and eventually causing browning and shedding of the leaves.

POINSETTIA, Euphorbia pulcherrima, in many instances reaching gigantic size, was occasionally observed seriously affected with the spot anthracnose disease, Sphaceloma poinsettiae Jenkins & Ruehle. The disease was exceedingly severe on certain plants, producing

many and large lesions on the stems and petioles, as well as spotting of the foliage and considerable hypertrophy. Often where the disease was severe, little or no new growth matured, and no floral bracts were produced.

SUGARCANE, Saccharum officinarum, appeared in most cases to be relatively free from common diseases. Leptosphaeria sacchari Breda de Haan, a leaf-spotting fungus, appeared on immature plants.

TOMATOES, Lycopersicon esculentum, were observed suffering mostly from the early blight fungus, Alternaria solani (Ell. & G. Martin) Sor. Little defoliation resulted. Certain plantings with mature green fruit showed mahogany rot, Phytophthora infestans (Mont.) D By., In several instances the fruits were 100 percent infected.

Nombre Vulgar del Huesped	Nombre Cientifico del Huesped	Causa Probable Nombre Cientifico	Enfermeded Nombre Vulgar
aguacate	Persea americana	Cercospora sp.	mancha de la hoja
aguacate	Persea americana	Sphaceloma persea	roña
almendro	Vitis labrusca	Guignardia bidwellii	mancha de la hoja
amate	Ficus aurea	Ophiodothella fici	mancha de alquitrán
amate	Ficus aurea	Cercospora bolleana	mancha cercospora
amate	Ficus aurea	Ramularia sp.	mancha de la hoja
amarilis	Amaryllis sp.	Cercospora amaryllidis	mancha cercospora
anona	Annona reticulata	Diplodia natalensis	pudrición del fruto
apio	Apium graveolens	Cercospora apii	mancha cercospora
apio	Apium graveolens	Rhizoctonia solani	mustia temprana
arroz	Oryza sativa	Helminthosporium oryzae	mancha de la hoja
berengena	Solanum melongena	Rhizoctonia solani	pudrición del suelo
berengena	Solanum melongena	Virus	mosaico
berengena	Solanum melongena	Diaporthe vexans	pudrición del suelo
café	Coffea arabica	Colletotrichum coffeicola	antracnosis
café	Coffea arabica	Pellicularia koleroga	mal de hilacha
café	Coffea arabica	Lichens	mancha de la hoja
café	Coffea arabica	Cercospora coffeicola	mancha cercospora
café	Coffea arabica	Phyllosticta sp.	mancha de la hoja
café	Coffea arabica	Omphalia florida	ojo de gallo
calabaza	Cucurbita pepo	Erysiphe cichoracearum	mildiú polvoriento
cana de azúcar	Saccharum officinarum	Leptosphaeria sacchari	mancha circular
chula	Vinca rosea	Corticium microsclerotia	mancha de la hoja
clavel	Hibiscus sp.	Rhizoctonia ramicola	mal de hilacha sedoso
clavel	Hibiscus sp.	Blakeslea trispora	moho
clavel	Hibiscus sp.	Puccinia malvacearum	roya
clavelón	Hibiscus malvaviscus	Rhizoctonia ramicola	mal de hilacha sedoso
crisantemo	Chrysanthemum hortorum	Septoria chrysanthemella	mancha de la hoja
crotalaria	Crotalaria vitelina	Virus	mosaico
croton	Croton sp.	Rhizoctonia ramicola	mal de hilacha sedoso
dalia	Dahliae pinnata	Entyloma dahliae	carbón
frijol terciopelo	Phaseolus vulgaris	Cercospora stizolobiae	mancha cercospora
frijol terciopelo	Phaseolus vulgaris	Bacterium sp.	mancha de la hoja
gladiola	Gladiolus hortulanus	Curvularia lunata	mancha curvularia
gladiola	Gladiolus hortulanus	Botrytis gladiolorum	mustia
graminias	Gramineae	Piricularia grisea	mancha de la hoja
granada	Punica granatum	Cercospora lythracearum	mancha cercospora
guineo	Musa paradisiaca	Cercospora musa	sigatoka
henequén	Agave sisalana	Capnodium sp.	fumagina
henequén	Agave latoriae	Septonema agaves	mancha de la hoja
higo	Ficus carica	Physopella fici	roya
higuerillo	Ricinus communis	Cercospora ricinella	mancha cercospora

jupiter	Lagerstroemia indica	Capnodium citri	fumagina
jupiter	Lagerstroemia indica	Rhizoctonia ramicola	mal de hilacha sedoso
jupiter	Lagerstroemia indica	Erysiphe lagerstroemiae	mildiu polvoriento
madreselva	---	Rhizoctonia ramicola	mal de hilacha sedoso
maicillo de escoba	Sorghum vulgare	Colletotrichum graminicola	antracnosis
maicillo de escoba	Sorghum vulgare	Helminthosporium turcicum	mancha de la hoja
maicillo de escoba	Sorghum vulgare	Puccinia purpurea	roya
maiz	Zea mays	Ustilago zeae	carbón
maiz	Zea mays	Helminthosporium turcicum	mancha de la hoja
maiz	Zea mays	Puccinia sorghi	roya
"Marigold"	Tagetes erecta	Septoria sp.	mancha de la hoja
mirasol	Helianthus annuus	Coleosporium helianthi	roya
naranja	Citrus sinensis	Sphaerostilbe aurantiicola	hongo de cabeza roja
naranja	Citrus sinensis	Elsinoë fawcettii	roña
orquides	Orchid sp.	Lichens	mancha de liquenes
papaya	Carica papaya	Asperisporium caricae	mancha negra de papaya
pascua	Euphorbia pulcherrimum	Virus	mosaico
pascua	Euphorbia pulcherrimum	Sphaceloma poinsettiae	roña
platanillo	Canna indica	Puccinia canniae	roya
pepino	Cucumis sativus	Corticium microsclerotia	mal de hilacha
pepino	Cucumis sativus	Phyllosticta cucurbitacearum	mancha de la hoja
pepino	Cucumis sativus	Pseudoperonospora cubensis	mildiú
pié de niño	Pedilanthus tithymoloides	Oidium sp.	mildiu polveriento
rábano	Raphanus sativus	Albugo candida	roya blanca
rosa	Rosa odorata	Cercospora rosicola	mancha cercospora
rosa	Rosa odorata	Diplocarpon rosae	mancha negra
rosa	Rosa odorata	Virus	mosaico
sorgo	Sorghum halepense	Gloeocercospora	mancha circular
sorgo	Sorghum halepense	Puccinia purpurea	roya
tomate	Lycopersicon esculentum	Alternaria tomato	mancha de cabeza de alfiler
tomate	Lycopersicon esculentum	Phytophthora infestans	mustia tardia
tomate	Lycopersicon esculentum	Alternaria solani	mustia temprana

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CENTRO NACIONAL DE AGRONOMIA, SANTA TECLA, EL SALVADOR

FUNDAMENTAL CONCEPTS IN THE DEVELOPMENT OF CONTROL MEASURES FOR SOUTHERN BLIGHT AND ROOT ROT ON PEANUTS[1]

Lytton W. Boyle[2]

Abstract

The distinction between necrotrophic and biotrophic parasites is reviewed. Aside from the semantic clarity obtained in the use of these terms, their practical application in development of control measures for certain diseases (Sclerotium rolfsii Sacc. and Rhizoctonia spp.) on peanuts is discussed.

INTRODUCTION

Life, whether in microbes or men, is at the mercy of whatever may control its supply of food. Although the concept of food control is very simple, we find its specific applications under many different names in widely separated and complicated fields of endeavor. In the ramifications of such a maze, as broad as life itself, this simple concept may often be obscured and opportunities for its application overlooked.

In the development and application of these tactics of food control to reduce certain plant diseases, the plant pathologist may encounter what appears to be a paradox. This appears when no distinction is made between the origin of the food supply in the live tissues of a host plant, and the nature of this same food as it becomes available to, and is assimilated by, allotrophic or pathogenic organisms located in the tissues of a host plant.

This distinction in the supply of food immediately available to a pathogenic organism has been reported numerous times and is a generally accepted phenomenon. In 1933, Link (4) reviewed this difference between the live and dead character of the food supply of respective parasites and proposed the terms "necrotrophic" (dead-food) and "biotrophic" (living-food) parasites to describe this difference. These terms have been ignored. Perhaps their value from a semantic point of view, as well as their application in the development of practicable control for certain diseases, has not feen fully appreciated. Such seemingly paradoxical situations and the value of such terms, are well illustrated in the case of southern blight (Sclerotium rolfsii Sacc.) and root rot (Rhizoctonia sp.) on peanuts.

FOOD CONTROL FROM A MACROSCOPIC POINT OF VIEW

The growth of the pathogenic fungus, Sclerotium rolfsii, is obviously at the expense of the living peanut plant. After such an observation of parasitism, it is often assumed that the vital factor in the live tissues of the peanut plant is also essential in the food of the (biotrophic) pathogenic fungus. It reasonably follows that the food of the fungus could be controlled by a scheme of crop rotation where peanuts follow several blight-resistant crops. Theoretically, the parasitic fungus would be starved, and reduced in numbers by other natural causes, while in the soil and separated from its host or vital food. Efforts to control southern blight by such methods have not been successful. It is evident that the fungus has not been sufficiently separated from its food by procedures that reasonably follow such macroscopic observations.

FOOD CONTROL FROM A MICROSCOPIC POINT OF VIEW

The microscopic relation between the hyphae of Scleratium rolfsii and their host cells has been fully described by Higgins (3). He observed that the fungus hyphae do not penetrate the living tissues of the host, and that death of the host cells is due to accumulation of oxalic acid, a metabolic product of the fungus. He also noted: "Apparently any plant with an epidermis easily permeable to oxalic acid solutions is susceptible to attack." "..... there is no occasion for the development of special adaptations between the parasite and host."

When the hyphae are limited to the dead tissues of the host plant, and no special adaptation is evident between the host and parasite, one may speculate that vitality acts as a barrier

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Table 1. The percentage of seed that emerge and remain healthy when sown with and without complementary necrotrophic media at respective levels in the soil.[a]

Depth of placement of medium in soil inches	Kind of media placed in soil: Corn Seeds sown No.	Corn Seedlings Emerge %	Corn Seedlings Healthy %	Cotton Seeds sown No.	Cotton Seedlings Emerge %	Cotton Seedlings Healthy %	Soybean Seeds sown No.	Soybean Seedlings Emerge %	Soybean Seedlings Healthy %	None Seeds sown No.	None Seedlings Emerge %	None Seedlings Healthy %
Soil naturally inoculated with *Rhizoctonia* spp.												
0-1	54	74.1	50.0	54	75.9	27.8	54	63.0	14.8			
1-2	54	83.3	37.0	54	59.3	37.0	54	68.5	16.7			
2-3	18	100.0	88.9	18	83.3	55.6	18	88.9	55.6			
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Soil inoculated with *Rhizoctonia* spp. and artificially[b] with *S. rolfsii*												
0-1	45	31.1	6.7	45	33.3	17.8	99	44.4	16.2			
1-2	45	42.2	28.9	45	22.2	11.1	45	31.1	17.8			
2-3	45	86.7	71.1	45	73.3	42.2	45	80.0	57.8			
0-3	—	---	---	--	---	---	--	---	---	81	91.4	87.7
0-1	27	51.9	00.0	27	66.7	7.4	27	51.9	3.7			
1-2	27	55.6	7.4	27	59.3	3.7	27	63.0	0.0			
2-3	18	94.4	44.4	18	66.7	27.8	18	88.9	5.6			
0-3	—	---	---	--	---	---	—	---		18	94.4	83.3

[a] Tests grown in greenhouse at 80°F. All percentages based on number of seed sown.

[b] Dry sclerotia separated from cultrue medium used to inoculate control plantings with no media in the soil. Culture medium of steamed oats incluedd with other inoculations.

Table 2. The number and condition of plants obtained from seeds sown in soil with complementary media placed at respective levels in the soil at respective intervals before seeding [a]

Placement of: medium and inoculum in soil [b] depth	days	Kind of media placed in soil: Corn: Stand of [c] plants	Corn: Location of lesions: Hypocotyl	Corn: Location of lesions: Roots: Secondary	Corn: Location of lesions: Roots: Tap	Cotton: Stand of plants	Cotton: Location of lesions: Hypocotyl	Cotton: Location of lesions: Roots: Secondary	Cotton: Location of lesions: Roots: Tap	Soybean: Stand of plants	Soybean: Location of lesions: Hypocotyl	Soybean: Location of lesions: Roots: Secondary	Soybean: Location of lesions: Roots: Tap
0-1"	0	18	3	12	6	13	8	7	0	8	0	7	1
	6	23	9	10	4	6	2	2	0	6	2	5	0
	14	26	8	12	4	16	6	9	0	8	5	6	1
	21	25	9	6	1	20	7	3	0	18	6	2	1
	27	22	1	0	0	25	3	0	0	26	4	5	0
	35	24	1	0	0	26	10	1	0	24	6	3	0
0-2"	0	19	2	5	4	13	2	4	2	5	0	0	4
	6	22	0	9	1	21	1	10	4	5	4	2	4
	14	24	3	5	7	23	4	7	0	16	4	3	8
	21	26	9	7	2	27	8	7	2	20	8	2	0
	27	24	1	0	0	22	1	6	0	23	2	2	2
	35	25	0	0	0	26	8	0	0	26	5	0	0
2-3"	0	17	6	7	14	20	1	4	11	20	2	3	17
	6	22	9	11	20	20	0	5	8	22	3	8	10
	14	24	0	1	22	22	0	10	11	12	3	4	2
	21	26	1	0	14	26	0	5	5	25	3	2	0
	27	25	1	0	0	23	4	0	0	25	5	0	0
	35	24	0	0	0	26	0	0	0	26	11	1	0

[a] Test grown in greenhouse at 80°F. Soil naturally inoculated with Rhizoctonia spp. and artificially inoculated with culture of S. rolfsii on steamed oats. Data collected 76 days after seeding.

[b] Medium of leaves and stalks were places with inocula in strata of soil at respectine depths (inches), and at respective numbers of days before seeding.

[c] Number of plants from 27 seeds sown in each planting.

rather than as an essential factor, in the food of this fungus. Although the food originates in the living plant, it is not available to the fungus except in crop refuse, dead weeds, or dead tissues of an otherwise live host plant. Such necrotrophic materials must therefore be the focal point in any attempt to control the pathogen by control of its food supply.

In the control of plant diseases, we are as yet without practicable therapeutic methods. Control must therefore be aimed at the prevention of infection. The food, or energy, used in establishment of a pathogenic relationship between a fungus and its host, may come from either of two sources. The reserve of food in a fungous spore or sclerotium may be sufficient to establish the pathogenic relationship. Food control then becomes the appalling task of destroying or removing all spores or sclerotia from the vicinity of the host. In the case of soil inhabiting pathogens, this in a very difficult, often impossible task. On the other hand, the reserve of food in the spore or sclerotium may not be sufficient to accomplish infection. Some medium, such as the above necrotrophic material is then needed as a complementary source of energy. Control is consequently designed either to destroy such media or to transfer them to an environment unfavorable to the pathogenic fungus.

Higgins has reported infection with S. rolfsii, both with and without a complementary medium (3). Infections were obtained with single sclerotia dropped beside tomato and soybean seedlings growing under artificial conditions in glass tubes with sterile tap water agar. Under more natural conditions in greenhouse culture, sclerotia remained dormant or inactive until stimulated by the accumulation of detritus from aphids. Under field conditions, it was noted that, "In soil containing large quantities of undecayed organic matter, every plant may be killed." It was further determined that the fungus is most active in a relatively warm (30-35°C), moist, well aerated environment(3).

GREENHOUSE TESTS OF INFECTION WITH AND WITHOUT A COMPLEMENTARY MEDIUM

Tests of the probability of infection without a complementary source of food or organic media were made in greenhouse culture (2). Soil was taken from a sandy bottom filled with a heavy cover of grass and some weeds. This soil was tossed through a 1/4-inch mesh, without special effort to work it through the screen. In this way, all organic matter except traces, such as a few short segments or fine roots, was removed. Some slightly discolored areas were sometimes found after close examination of the secondary roots of peanut plants growing in such soil. These were few and of questionable origin. Otherwise the root systems appeared perfectly healthy. The root systems of a second planting of peanuts were found to be equally free from infection if the roots of the preceding planting had been removed from the soil. However, lesions of Rhizoctomia sp. were frequent on the roots of the second planting if the tops only of the first planting had been removed and the root systems left undisturbed in the soil. With the dead roots of the first crop as a complemenatry source of food or energy, the natural inoculum in the soil became effective with the second planting.

The effect of various sorts of organic matter in such soil was next tested. Plant materials such as the stems and leaves were gathered from soybean, cotton, and corn fields. Mature plants were collected in the early fall before becoming lodged or weathered. Care was taken to cut all plants several inches above the soil level and to avoid any stalks or leaves that appeared to have made any contact with the soil. When such materials were added in the upper two or three inches of soil, the peanut plants became severely infected with a species of Rhizoctonia (Table 1).

As in the case of Rhizoctonia sp., the sclerotia of S. rolfsii were innocuous without a complementary supply of organic materials in the soil. Sclerotia used for the tests were cleaned from dried cultures grown on autoclaved oats by screening over an air draft. However, when organic trash was added to the upper inches of soil with such sclerotia, the emergence of peanut plants was greatly reduced and many severe lesions were found on the hypocotyls and roots of plants that did emerge (Table 1).

The incidence and severity of infection obtained with complementary media varies according to several factors (Table 2). These are the kind of organic matter, depth of its placement in the soil, and the length of time it has remained in the soil before peanuts are planted. The effect of leaves and stems from a respective crop varied greatly between samples. However, both the incidence and the severity of disease were generally greater with soybeans, followed in order by cotton and corn. The effect of depth of placement is primarily ecological, due to temperature and aeration. In consideration of the time factor, two possibilities appear. First, it is assumed that the greater part of the nutrients in the organic matter is rapidly consumed by biological activity in such relatively warm soil. In this way, the amount of nutrient available to the pathogen might be reduced to less than the required amount for establishing infection of

host tissues. This priority in access to available food, or somewhat competitive relationship, has been suspected in many previous tests not designed to allow its accurate measurement. It has been consistently observed under a variety of conditions both in the field and in greenhouse plantings. On the other hand, the possibility of antagonistic or antibiotic relationships between the soil flora and pathogens can not be completely ruled out (5). However, in the development of a practicable method of control, there is little assurance that either possibility would be consistently effective under a wide range of conditions.

In the case of southern blight and root rot on peanuts, good control has been obtained by cultural practices, when these are carried out with precision (1). These practices are designed, first, to bury all necrotrophic media in a depth of soil where growth of the pathogen is checked by ecological factors; and, second, to prevent accumulation of such media about the base of the plant during the crop season. Such media accumulate where insect damage is severe, and where leaves are smothered by cultivation or dropped because of leafspots.

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GEORGIA EXPERIMENT STATION, EXPERIMENT, GEORGIA, AND VEGETABLE CROPS SECTION, HORTICULTURAL CROPS RESEARCH BRANCH, AGRICULTURAL RESEARCH SERVICE COOPERATING.

THE EXPORTATION OF PLANT MATERIALS[1]

W. H. Wheeler[2]

On occasion American plant pathologists find it desirable to export plant materials to scientists or other persons in foreign countries. In many instances, however, they are unaware of the plant quarantine restrictions that may govern the entry of such plant materials into the country of destination.

An examination of the pertinent regulations indicates that certain countries require the importer to hold a permit authorizing entry of the plant material. Other regulations may also require that the shipment shall be accompanied by a phytosanitary certificate indicating freedom from insect pests and plant diseases. Some countries require that such certificates shall be issued by the Federal Government but others will accept those prepared by the several states. It should also be noted that many countries prohibit the entry of certain things altogether, or only admit them under a specially issued permit for scientific or experimental purposes.

Among the things that may be prohibited entry into certain countries are cultures of disease-producing organisms. Other items will include seeds, plants, bulbs, etc., that may carry certain serious pests or diseases not recorded in the country of destination.

Under authority of the Department of Agriculture Organic Act of 1944, the Plant Quarantine Branch of that Department has summarized all plant quarantine regulations known to have been issued by the countries of the world. The purpose in doing so has been to render service to American exporters which will enable them to meet the foreign phytosanitary requirements and avoid rejection of their shipments upon arrival at foreign ports.

It should be pointed out that such service can be effectively rendered if it is requested before the shipment is dispatched to the foreign country, since the acutal goods to be covered by the certificate must be examined. Added to that requirement is a newer one being enforced by certain countries which specifies that the material to be shipped must have been examined during the growing season as well as at the time of export. Thus, it behooves the prospective shipper of plant materials to inquire into the plant quarantine requirements of the foreign country of destination so that he can secure a certificate that will cover the growing season inspection as well, if it is needed.

It often develops that the person in the foreign country who has requested plant material from this country is completely unaware of the plant quarantine requirements of his own country. Therefore, he may ask for some prohibited item such as sweet corn seed when the regulations of the country state it shall not be imported except under a certificate stating that Stewart's disease does not exist in the area where the seed were produced. Such a certificate can not be furnished by several of the corn countries, but neither the shipper nor the receiver may be aware of that regulation unless an inquiry is made. Therefore, American shippers should make inquiry regarding the plant quarantine regulations of the receiving country, unless the importer has furnished that specific information. To get such information the shipper in this country should address his inquiry to the Plant Quarantine Branch, Agricultural Research Service, U. S. Department of Agriculture, Washington 25, D. C., or to one of the offices of the Plant Quarantine Branch located in the principal ports of entry on both coasts and on the Canadian and Mexican Borders.

PLANT QUARANTINE BRANCH, AGRICULTURAL RESEARCH SERVICE,
UNITED STATES DEPARTMENT OF AGRICULTURE

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[1]Prepared on behalf of the Committee on Seed and Plant Material Certification, American Phytopathological Society.

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NEW AND UNUSUAL RECORDS OF PLANT DISEASES

STEM ROT OF POTATO[1]

by Robert D. Shealy[2]

A stem rot of potato was noted in Ohio in August 1955 by the author while inspecting potato fields for the Ohio Agricultural Extension Service. Symptoms include a soft rot of potato stems which developed the latter part of the growing season. No tuber rot was found in the field which could be correlated with this disease. A bacterium was isolated repeatedly from diseased stems and incited the disease on plants in the greenhouse. Tubers were planted in soil collected in early March 1956 from fields in which the disease had been severe in 1955. Symptoms developed on plants growing in this soil in the greenhouse. The bacterium was pathogenic to many members of the Solanaceae.

The bacterium is a short rod, motile, and Gram-negative. Acid is produced from many sugars, including lactose, but gas is not produced in any of the media thus far tested. The morphology, cultural characteristics, and biochemical reactions are not similar to those of any described bacterial pathogen of potato. Further studies of the disease and pathogen are in progress.

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SUSCEPTIBILITY OF GOMPHRENA TO TOMATO RINGSPOT VIRUS

By Philip Brierley[1]

On using Gomphrena globosa as an index plant for detection of hydrangea viruses, we were surprised to find that the tomato ringspot virus produces symtoms in it. Although Gomphrena is known to be susceptible to many other viruses, we find no record of it as a host of tomato ringspot virus. Gomphrena is as sensitive as tobacco in detecting tomato ringspot virus in florists' hydrangea. Solid gray spots appear after 6 to 10 days, becoming red-bordered after 5 to 6 days more, and systemic with gray necrotic spots and sometimes tip killing after 28 days. The virus is sub-transferable from Gomphrena to Gomphrena, and to tobacco in high titer. On the other hand, we have failed to transmit it from tobacco to Gomphrena in 5 trials, even when healthy hydrangea leaves were ground with the tobacco inoculum. Similarly tomato-ringspot virus failed to transfer from infected tomato or snapdragon to Gomphrena.

Gomphrena is thus doubly useful as an index plant for hydrangeas, detecting both hydrangea and tomato ringspot viruses. The latter is easily separable from the former through its capacity to invade Gomphrena systemically and to infect tobacco.

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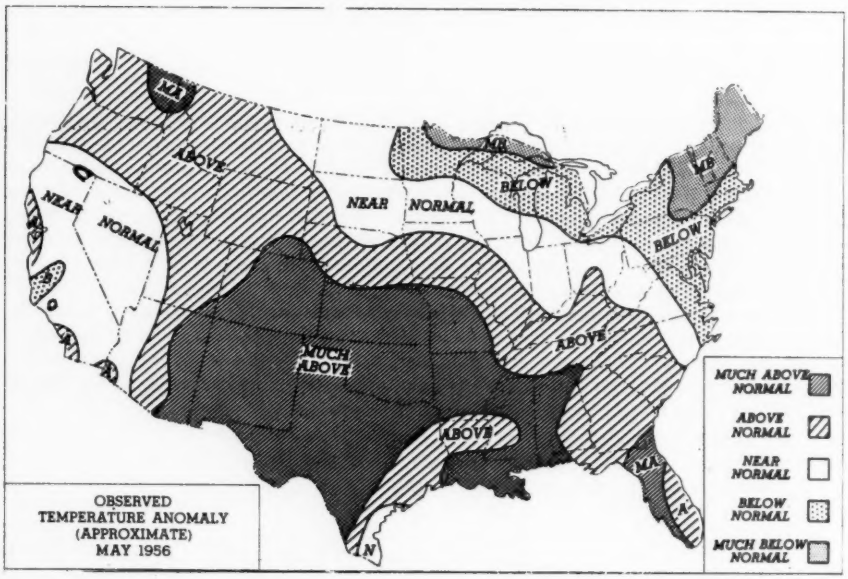

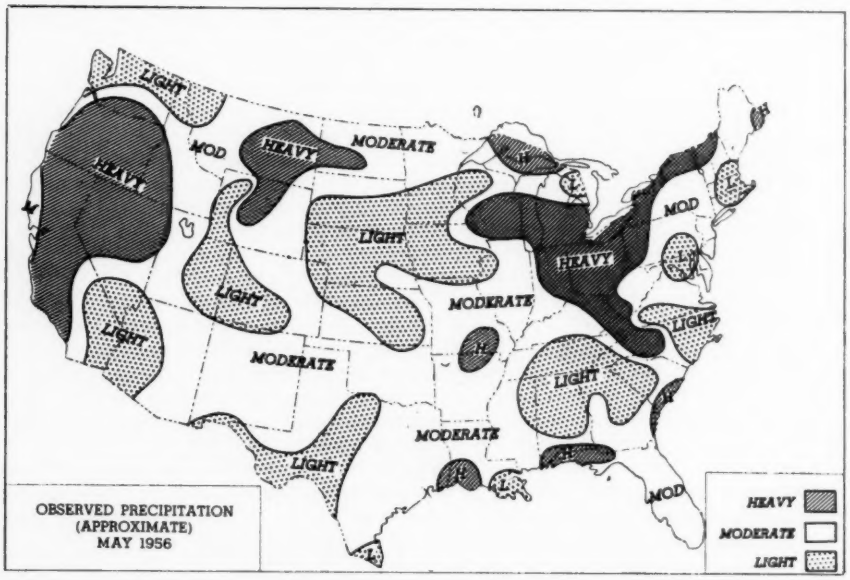

The terms used in the accompanying maps, which define the ranges of temperature and precipitation, are numerical class limits. These are based on a statistical analysis of past records through which is determined the normal frequency of occurrence of temperatures and precipitation at various times of the year for different locations. For temperature the classes above, below, and near normal are so defined that they each normally occur one-fourth of the time; much above and much below normal, one-eighth of the time Precipitation is depicted in terms of light, moderate, and heavy, each class normally occurring one-third of the time and thereby having equal probability of occurrence. These maps graphically represent only the general trends and give the country's weather picture at a glance. For quantitative studies, where monthly mean temperatures and actual precipitation records are needed for a given time and place, other publications of the Weather Bureau should be consulted. P. R. M.

MANUSCRIPTS FOR THE PLANT DISEASE REPORTER

(See also PDR 35: 121-122; 36: 214, 494)

(1) GENERAL: The Reporter page measures 9 inches long with the heading or 8 3/4 inches for the text part, by 6 inches wide. The copy is typed on a larger page, 11 1/4 inches of text or 12 inches overall in length by 8 inches in width, and reduced 25 percent in the photographic process of reproduction. Illustrations or tables larger in either dimension will take a correspondingly greater reduction. Only one size of type is available for text, footnotes, or tables.

(2) MANUSCRIPTS should be the original ribbon copy, not carbons, clearly typed and double-spaced throughout, including tables, footnotes, and bibliographies. (Note -- only one copy is needed.) Footnotes should be typed at the bottom of the page.

(3) ABSTRACTS are requested for all except very short articles. (PDR 38: 316).

(4) CAUSES OF DISEASES should be named. For bacteria, fungi, nematodes, etc., give the Latin name of the organism; for viruses either or both the accepted common name of the virus or a Latin name if you prefer it and there is one; for non-parasitic diseases state the causal factor if it is known. If the cause of a disease has not been determined say so. (PDR 36: 494).

(5) LITERATURE REFERENCES should be given in alphabetical order and numbered for citation in the text. We follow the AIBS suggestion of placing the year of publication after the author's name. Please check your titles carefully since we cannot do it for you. Be sure that text citations and bibliography agree; that foreign-language references are correct; that number or month is cited for periodicals that are not paged consecutively throughout the volume.

(6) NAMES OF FUNGICIDES should be given according to the suggestions that will be published in Phytopathology before long, and later repeated in the Reporter. In the meantime see PDR 35: 122.

(7) ILLUSTRATIONS should be sent to us unmounted. To prevent mistakes, write figure numbers on the back, and mark the top of each print when necessary. A sketch can show a preferred arrangement but please keep in mind page size, shape, and standard reduction (see above under General), and remember that figure titles and legends are part of the page. Lettering should be clear and large enough to be legible after reducing. Drawings, maps, and graphs can be photographs or originals, but should be finished and ready for reproduction, not just sketches.

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